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B-cell translocation gene 2 promotes hepatic hepcidin production via induction of Yin Yang 1

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ABSTRACT

Hepcidin is a peptide hormone secreted in the liver and plays a key role in maintaining iron homeostasis. Here, we demonstrate that B-cell translocation gene 2 (BTG2) is a key player in hepatic hepcidin regulation via induction of Yin Yang 1 (YY1). Hepatic hepcidin gene expression significantly enhanced by fasting states and glucagon exposure led to induction of gluconeogenic gene expression, and elevated serum hepcidin production in mice. Notably, overexpression of BTG2 using adenoviral system (Ad-BTG2) significantly elevated serum hepcidin levels via a significant induction of YY1 gene transcription. Immunoprecipitation studies demonstrated that BTG2 physically interacted with YY1 and recruited on the hepcidin gene promoter. Finally, ablation of hepatic BTG2 gene by gene silencing markedly attenuated the elevation of serum hepcidin production along with YY1 and hepcidin mRNA expression in fasting state. Likewise, forskolin (FSK)-stimulated hepcidin promoter activity was dramatically disrupted by endogenous BTG2 knockdown. Overall, our current study provides a novel molecular mechanism of BTG2-mediated induction of hepcidin gene expression, thereby contributing to a better understanding of the hepatic hepcidin production involved in iron homeostasis.

1. Introduction

Hepcidin is a small peptide hormone mainly produced by hepatocytes in the liver that plays a key role in maintaining mammalian iron homeostasis. It represses dietary iron absorption in the enterocytes, recycling by macrophages, and the release of storage iron from hepatocytes by binding and/or triggering the internalization and degradation of ferroportin, which causes a reduction in extracellular iron concentration [1]. Hepatic hepcidin production is elevated under various physiological conditions, including infection, inflammation, and severe iron overload, whereas this phenomenon is attenuated by hypoxia, iron-deficiency anemia, erythropoietin, growth factors [1,2]. Moreover, hepcidin gene expression is increased in response to stimulatory signals such as bone morphogenetic proteins (BMP)/SMAD axis, interleukin (IL-6)/signal transducer and activator of transcription 3 (STAT3), cAMP responsive element binding protein H (CREBH), whereas it is suppressed by hypoxia inducible factor (HIF), growth differentiation factor (GDF15), twisted gastrulation protein homolog 1 (TWSG1) [1–3]. A recent report has shown that gluconeogenic signals control mammalian iron homeostasis via induction of hepcidin in starving mice [4]. Although gluconeogenic signals regulate hepcidin production in mice, the basic molecular mechanism of hepatic hepcidin regulation caused by gluconeogenic system has not been fully investigated.

Yin Yang 1 (YY1) is a multifunctional transcriptional factor of the polycomb group protein family, and exerts as a transcriptional repressor and activator by binding to consensus sequences (CCATNTT). YY1 is widely expressed in diverse tissues and involved in embryogenesis, replication, cellular proliferation, and

Abbreviations: Ad, adenovirus; BTG2, b-cell translocation gene 2; ChIP, chromatin immunoprecipitation; Dis, distal region; FSK, forskolin; GFP, green fluorescent protein; GLU, glucagon; IB, immunoblot; IP, immunoprecipitation; MT, mutant form; Pro, proximal region; qPCR, quantitative polymerase chain reaction; shRNA, short hairpin RNA; siRNA, small interfering RNA; WT, wild type; YY1, Yin Yang 1.

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differentiation [5]. Moreover, YY1 also associates with a diverse protein regulating cellular proliferation and apoptosis, such as p53, murine double minute 2 (Mdm2), retinoblastoma (Rb), histone deacetylase complex (HDAC), c-Myc, and other transcription factors [6]. Recently, YY1 promotes gluconeogenesis and steatosis by controlling glucocorticoid receptor and farnesoid X receptor in the livers of mice [7,8]. However, the link between YY1 and hepcidin production involved in the regulation of iron homeostasis remains unexplored.

BTG2 (B-cell translocation gene 2) is a member of BTG/Tob gene family of anti-proliferative genes that acts as a key regulator of the cellular machinery, including cell growth, differentiation, death, and survival [9]. Especially, BTG2 is stimulated by hypoxia, genotoxic stress, metabolic changes, retinoic acid, and attenuated by insulin, estrogen, growth factors [10]. Previously, we have elucidated that BTG2, a transcriptional co-activator, regulates the transcription of several target genes, such as gluconeogenic genes, insulin [11,12]. However, the potential role of BTG2 in the regulation of hepatic hepcidin production and its subsequent role in maintaining iron homeostasis have not been investigated.

In the current study, we demonstrate that BTG2 acts as a novel partner of hepatic hepcidin regulation and reveals a basic molecular mechanism that links to YY1 by driving hepatic hepcidin production in gluconeogenic pathway.

2. Materials and methods

2.1. Animal experiments

Male 8-week old C57BL/6 mice (Samtako, Osan, Republic of Korea) were used for this experiment, as below described [13]. For studies in the fasting and feeding conditions, mice were fed *ad libitum* and fasted for 12 h. For glucagon stimulation experiments, glucagon (Sigma–Aldrich, St. Louis, MO, USA) was injected intraperitoneally into mice at a dose of 10 μg/kg body weight for 6 h. For overexpression of BTG2, WT mice were infected with adenoviral vector expressing BTG2 (1.10^9 plaque-forming units, pfu) by tail vein injection. For ablation of BTG2 gene, WT mice were intravenously injected with lentivirus short hairpin BTG2 (sh BTG2, a single dose of 1.10^9 transducing units (TU) per ml). Livers from fast, adenovirus- and lentivirus-infected mice were utilized in the preparation of total RNA and/or protein. All animal experiments were performed in accordance with the rules and regulations of the Institutional Animal Use and Care Committee (IAUCC), Keimyung University School of Medicine.

2.2. Measurement of serum hepcidin

Serum hepcidin level was measured using an enzyme-linked immunosorbent assay kit (Uscn Life Science Inc., Hubei, China) in accordance with the manufacturer's instructions, as previously described [14].

2.3. Plasmids and DNA constructs

Mouse hepcidin gene promoter was kind gifted by Dr. Hueng-Sik Choi (Chonnam National University, Gwangju, Republic of Korea) [14]. The YY1 response element-mutated hepcidin promoter was generated using a Site-Directed Mutagenesis kit (Stratagene, La Jolla, CA, USA) and the following primers: forward, 5′-GAGTAACAGTTTTACTGAAGGCAC-3′, and reverse, 5′-GTGCCTTCAGTAAAACTGTTACTC-3′. Expression vector for BTG2 was described previously [13] and pCMVSPORT6-YY1 plasmids was kindly provided by Dr. Gap Ryol Lee (Sogang University, Seoul, Republic of Korea). All constructs were confirmed by DNA sequencing.

2.4. Cell culture and transient transfection assays

AML-12 cells (immortalized mouse hepatocytes) were cultured in DMEM/F-12 medium (Gibco-BRL, Grand Island, NY, USA) supplemented with 10% fetal bovine serum (Gibco-BRL), insulin-transferrin-selenium (Gibco-BRL), dexamethasone (40 ng/ml, Sigma–Aldrich), and antibiotics in a humidified atmosphere containing 5% CO_2 at 37 °C [15]. Transient transfections were carried out as previously described [13,15].

2.5. Isolation and culture of primary mouse hepatocytes

Mouse primary hepatocytes were performed from the livers of 8-week-old male mice (Samtako), as previously described [13].

2.6. Preparation of recombinant adenovirus and siRNA experiments

Adenoviruses encoding full length BTG2 (Ad-BTG2), green fluorescent protein (GFP) and lentiviral BTG2-targeted short hairpin RNA (sh RNA) were described previously [13]. The siRNAs for BTG2 (si Scram and si BTG2) were chemically synthesized (Bioneer Research, Seoul, Republic of Korea), and transfected according to the manufacturer's instructions, as previously described [11].

2.7. RNA isolation and analysis

Total RNAs were isolated from liver using TRIzol reagent (Invitrogen, Carlsbad, CA, USA) as previously described [13,15]. Quantitative polymerase chain reaction (qPCR) analysis were conducted using primers of BTG2, Nur77, YY1, and hepcidin as described previously [11,13,14]. The expression of all transcripts was normalized to ribosomal L32 expression.

2.8. Co-immunoprecipitation assay

Total protein isolated from mouse liver was immunoprecipitated with antibodies against BTG2 and YY1 (Santa Cruz Biotechnology, Santa Cruz, CA, USA), and then blotted with these antibodies [13]. Signals were detected with ECL-Plus Western blot detection kit (Amersham Bioscience, Piscataway, NJ, USA).

2.9. Chromatin immunoprecipitation (ChIP) assay

The ChIP assay was performed as described previously [11,13]. Briefly, after 36 h infection with Ad-BTG2 in mouse primary hepatocytes, cells were treated with FSK (10 μM) for 6 h. The cells were subsequently harvested, and ChIP assay was performed with anti-YY1. After purification, DNA samples were quantified by PCR with two pairs of primers for the proximal (−450/−250 bp) and distal (−1900/−1700 bp) region of the hepcidin gene promoter. The specific primers used for PCR are as follows: proximal, forward 5′-ATATGGTCTTCACAGTGGTC-3′ and reverse 5′-CAGTGATTGGATTGGTGAGT-3′; distal, forward 5′-TTGGGTAGGGTTCCTAGGGT-3′ and reverse 5′-CGT-GGTACAGAGGGAGGGCT-3′.

2.10. Statistical analysis

Results are expressed as means (±S.E.M.). Analysis of variance was employed to determine significant differences as detected by Student's *t* tests and/or ANOVA methods using prism program. Statistical significance was considered at $P < 0.05$.

3. Results

3.1. Hepatic hepcidin production is elevated by gluconeogenic signals

A previous report has shown that gluconeogenic signals regulate hepcidin metabolism via the peroxisome proliferator-activated receptor γ coativator-1α (PGC-1α) and cAMP response element-binding protein H (CREBH)-dependent pathway in starving mice [4]. Based on this finding, we investigated the physiological link between gluconeogenic partners and hepatic hepcidin regulation. The mRNA and protein levels of hepcidin were significantly enhanced under fasting condition along with the expected induction of BTG2 and YY1, which contribute to the hepatic gluconeogenesis (Fig. 1A). Interestingly, serum hepcidin levels were also elevated under fasting condition, when compared with that of fed state (Fig. 1B). Next, we examined the effect of gluconeogenic signal on the regulation of hepcidin gene expression in the livers of mice. As shown in Fig. 1C and D, treatment of glucagon (GLU) significantly increased the mRNA levels of hepcidin, BTG2, and YY1 (Fig. 1C). Likewise, GLU exposure showed elevated the protein level of BTG2 and YY1 as well as serum hepcidin level compared to controls (Fig. 1D). Overall, these results strongly suggest a novel potential role between BTG2 and hepatic hepcidin production caused by gluconeogenic signals.

3.2. BTG2 elevates hepatic hepcidin production

To elucidate the crucial role of BTG2 as a positive regulator of hepatic hepcidin production, we performed adenoviral delivery of BTG2 (Ad-BTG2) and GFP (Ad-GFP) in mouse liver. Ad-BTG2 transduction significantly elevated serum hepcidin levels compared to controls (Fig. 2A). Ad-BTG2 was effectively delivered to C57BL/6 (WT) mice via tail vein injection and successfully overexpressed in the liver. Consistent with the change of YY1 and hepcidin mRNAs, protein levels were also increased in Ad-BTG2-infected mouse liver compared to Ad-GFP controls (Fig. 2B). To further confirm the transcriptional regulation of hepcidin in response to BTG2, we examined hepcidin promoter activity in AML-12 cells. Remarkably, BTG2 significantly increased the transcriptional activity of hepcidin in a dose-dependent manner, and this phenomenon was further enhanced by the co-transfection of YY1 (Fig. 2C). Collectively, these observations demonstrate that BTG2 may act as a key regulator of hepcidin production in the liver.

3.3. BTG2 physically interacts with YY1 and mediates YY1 recruitment on the hepcidin gene promoter

To explore the connection between BTG2 and YY1 in the regulation of hepcidin gene expression, we performed Co-immunoprecipitation (Co-IP) assays using BTG2-and YY1-specific antibodies. Endogenous BTG2 was highly immunoprecipitated with YY1 protein in fasted mouse liver than fed liver (Fig. 3A). We further evaluated the functional significance of the YY1-binding site on the hepcidin gene promoter using point mutation experiments. Like BTG2, YY1 significantly enhanced hepcidin gene promoter activity in a dose-dependent manner, and this phenomenon was totally absent in the YY1-binding site-mutated hepcidin gene promoter (Fig. 3B). These results display that BTG2 physically interacts with YY1. To further conform whether BTG2 affects YY1 occupancy on the hepcidin gene promoter, we performed chromatin immunoprecipitation (ChIP) assay in mouse primary hepatocytes. As shown in Fig. 3C, the endogenous YY1 recruitment to the proximal region in response to forskolin (FSK) treatment was significantly promoted by Ad-BTG2 compared to controls. However, the nonspecific distal region of the hepcidin gene promoter was unable to recruit YY1 under all conditions. Taken together, these results strongly suggest that BTG2 enhances hepcidin gene transcription via the recruitment of YY1.

3.4. Elevation of hepatic hepcidin production by gluconeogenic signals is mediated by BTG2

We revealed the potential role of BTG2 in hepcidin production caused by gluconeogenic system in the livers of mouse using knockdown system of BTG2 (lentivirus-sh BTG2). Fasting state

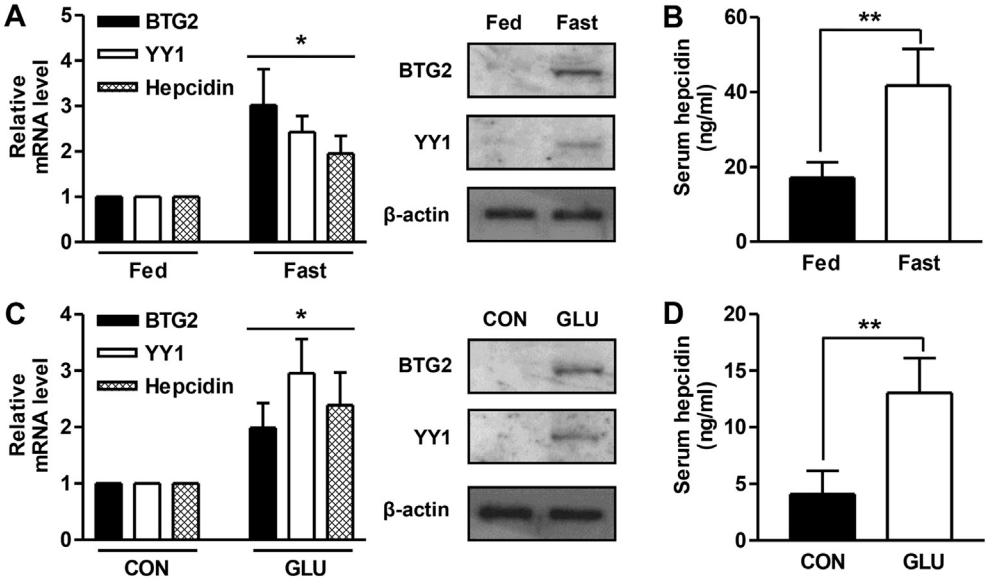

Fig. 1. Hepatic hepcidin gene expression is induced by fasting condition and glucagon exposure. (A) C57BL6 (WT) mice (n = 7) were fed *ad libitum* and fasted for 12 h. Total RNAs extracted from livers were measured to qPCR analysis with indicated primers. (B) Serum hepcidin level in WT mice. (C) WT mice were injected intraperitoneally with glucagon (10 μg/kg body weight) for 6 h and then measured by qPCR with the indicated primers. (D) Serum hepcidin level in the indicated mice. $^{*}P < 0.05$, $^{**}P < 0.01$ vs. fed mice or untreated control mice.

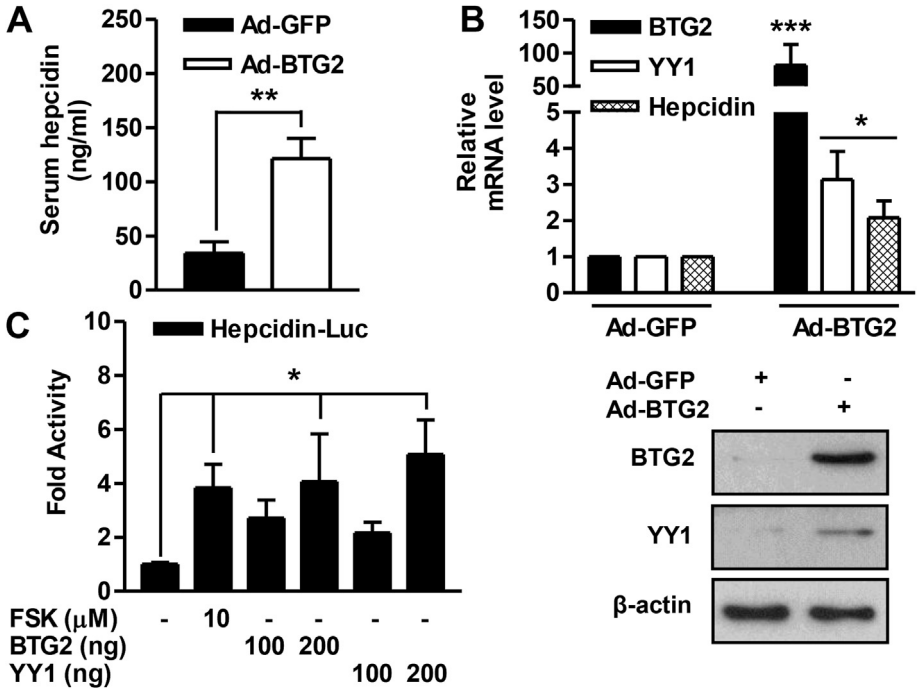

Fig. 2. BTG2 promotes hepcidin production. (A) Serum hepcidin level in WT mice infected with Ad-GFP and Ad-BTG2 for 5 days. (B) Gene expression of hepcidin and other transcription factors were measured to qPCR analysis in mouse liver (n = 7). (C) AML12 cells were transiently transfected with BTG2, YY1 and the indicated reporter gene, and then treated with FSK for 6 h $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$ vs. Ad-GFP or untreated control.

significantly elevated serum hepcidin levels and it was markedly disrupted by silencing of BTG2 (Fig. 4A). Moreover, elevation of YY1 and hepcidin mRNA levels in response to fasting state were dramatically attenuated by endogenous BTG2 knockdown compared with controls (Fig. 4B). We further investigated whether FSK-stimulated hepcidin promoter activity is mediated by BTG2 in hepatocytes. As expected, the transcriptional activity of hepcidin was significantly increased by FSK stimulation, and this effect was strikingly abolished by silencing of BTG2 (Fig. 4C). Overall, these findings strongly suggest that elevation of hepatic hepcidin production caused by gluconeogenic system is BTG2 dependent.

4. Discussion

In this study, we have demonstrated that glucongeogenic signals enhanced hepcidin gene expression, and BTG2 promoted hepatic hepcidin production by upregulating YY1. Based on these findings, we propose that glucagon-BTG2-YY1 signaling pathway may provide a molecular mechanism underlying regulation of hepcidin gene expression as well as a major modulator of hepatic hepcidin production.

Previous studies have represented that gluconeogenic signals, known as key regulators of hepatic gluconeogenesis, modulate hepcidin gene expression in mice and human volunteers. These studies also suggested that glucagon might mediate the production of hepatic hepcidin, which involves in iron homeostasis [4,16,17]. Moreover, the increase of BTG2 caused by glucagon resulted in the elevation of hepatic gluconeogenesis in mice [13]. However, there is no evidence of involvement of BTG2 in driving a hepatic hepcidin gene expression and/or affecting hepcidin production. Our current results revealed that elevation of BTG2 by gluconeogenic signals control hepatic hepcidin production by promoting YY1. Fasting state and glucagon exposure significantly increased BTG2, YY1, and hepcidin mRNA levels in the livers of mice along with a significant elevation of serum hepcidin level (Fig. 1) and this phenomenon was dramatically disrupted by silencing of BTG2 (Fig. 4). Moreover, overexpression of BTG2 using adenoviral system significantly elevated hepcidin production by promoting YY1 (Fig. 2). Overall, our current findings suggest that BTG2 play a key role in driving glucagon-stimulated hepatic hepcidin production through YY1 induction.

Recently, YY1 is characterized to regulate expression of several target genes through recruitment of co-activator PGC-1α and steroid receptor co-activator 1 (SRC-1) [7,18]. Based on these findings, we explored the fundamental molecular mechanism of glucagon-induced hepcidin gene expression is mediated by BTG2-YY1 axis in hepatocytes. First, physical interaction of BTG2 and YY1 was observed in livers of fasted-mice. Second, endogenous YY1 binds directly to a proximal site on the hepatic hepcidin gene promoter under both gluconeogenic signal exposure and Ad-BTG2 transduction, and the transcriptional activity of hepcidin was also enhanced by BTG2 and YY1 in hepatocytes (Fig. 3). Overall, our findings strongly provide a basic molecular mechanism between hepatic hepcidin gene regulation and BTG2-YY1 signaling network. However, we cannot exclude the possibility that the detailed molecular mechanism of connection between hepatic hepcidin production and BTG2-YY1 axis may rely on unknown mechanism of co-regulator competition or recruitment, unrevealed transcription factors, protein stability and other signal molecules to drive the regulation of hepatic hepcidin gene expression.

In conclusion, our results demonstrate that hepcidin is a novel target of BTG2 and BTG2 enhances hepatic hepcidin production through induction of YY1. Moreover, these findings suggest that elevation of BTG2 in response to gluconeogenic signals regulate iron homeostasis by promoting YY1-mediated hepatic hepcidin production. Therefore, a novel molecular mechanism involved in hepcidin production caused by BTG2 may provide an attractive strategy for the development of novel therapeutic agents to treat iron dysfunction like hereditary hemochromatosis and anemia.

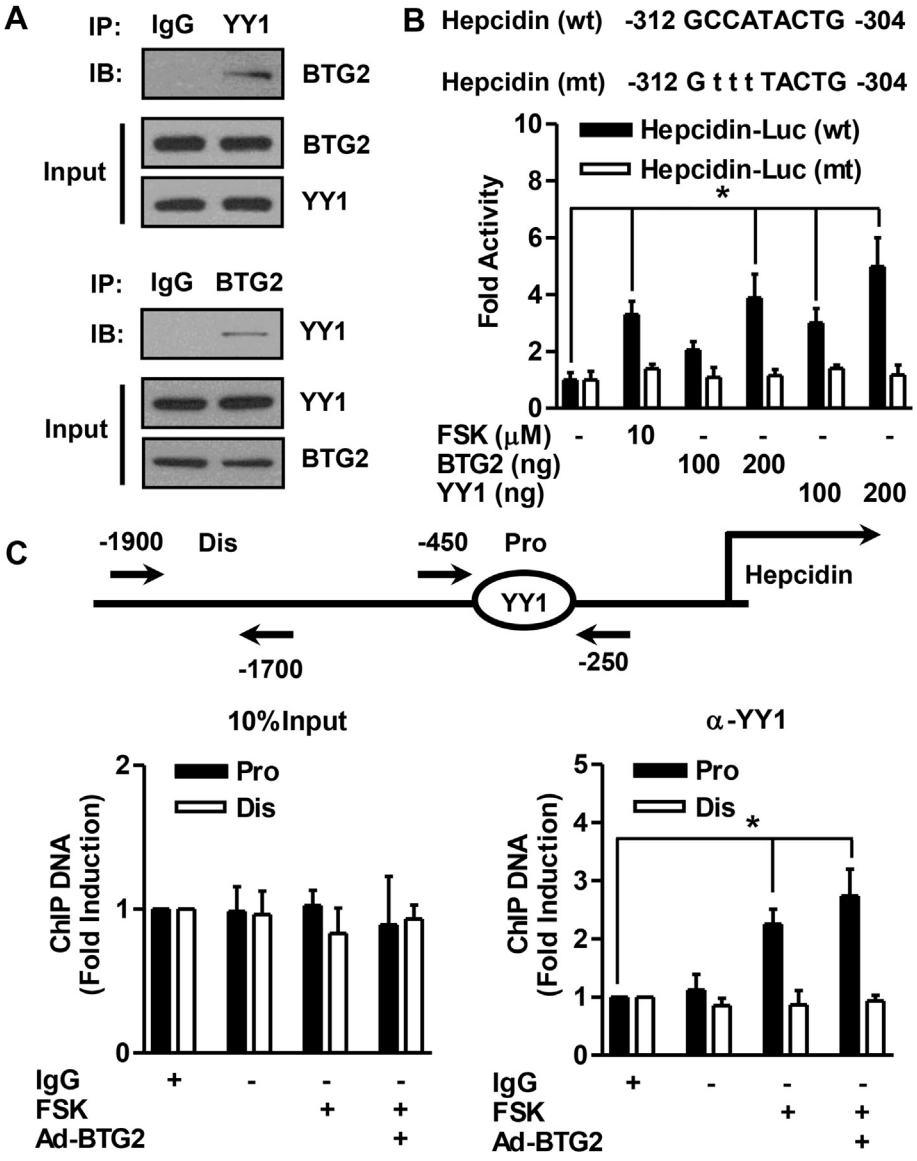

Fig. 3. Interaction of between BTG2 and YY1. (A) Functional association between BTG2 and YY1 in the liver of fasted-mice for 12 h. Protein extract from livers was immunoprecipitated with YY1 and immunoblotted with BTG2 antibody. (B) Schematic diagrams of wild-type (wt) and mutant form of the hepcidin gene promoter (mt) constructs from −312 to −304 bp as indicated conditions. AML12 cells were transiently transfected with BTG2, YY1 and the indicated constructs, and then treated with FSK for 6 h. (C) Chromatin immunoprecipitation (ChIP) assay showing the recruitment of YY1 on the hepcidin gene promoter. Mouse primary hepatocytes were infected with Ad-BTG2 for 24 h, and then treated with FSK for 6 h, respectively. Input displays 10% of purified DNA. Soluble chromatin was immunoprecipitated with anti-YY1 antibody or IgG as indicated. Purified DNA samples were employed for qPCR with primers binding to the specific proximal (Pro) and nonspecific distal (Dis) region on the hepcidin gene promoter. $^{*}P < 0.05$ vs. untreated control.

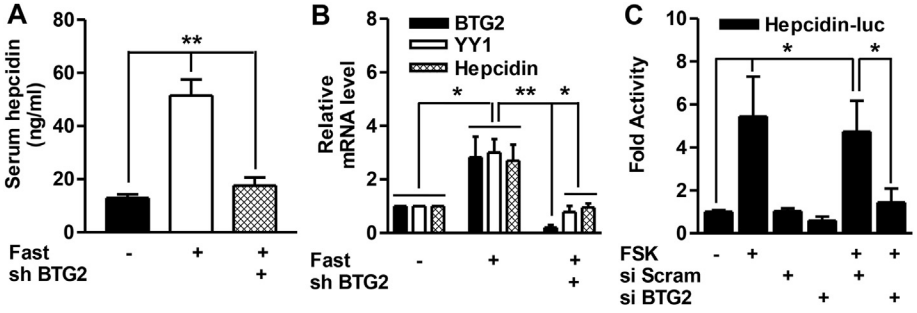

Fig. 4. Induction of hepatic hepcidin production caused by glucagon signals is BTG2 dependent. (A) Five days after sh BTG2 infection, mice were fasted for 12 h and measured for serum hepcidin levels. (B) Total RNAs extracted from mouse liver were analyzed by qPCR with the indicated primers. (C) AML12 cells were transfected with the si BTG2 and si Scram. After transfection for 36 h, cells were transfected with the indicated reporter gene, and then treated with FSK for 6 h $^{*}P < 0.05$, $^{**}P < 0.01$ vs. untreated control or fast mice or FSK-treated cells.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgments

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References

[1] D. Meynard, J.L. Babitt, H.Y. Lin, The liver: conductor of systemic iron balance, Blood 123 (2014) 168–176.
[2] A. Pietrangelo, Hepcidin in human iron disorders: therapeutic implications, J. Hepatol. 54 (2011) 173–181.
[3] T. Ganz, E. Nemeth, Hepcidin and iron homeostasis, Biochim. Biophys. Acta 1823 (2012) 1434–1443.
[4] C. Vecchi, G. Montosi, C. Garuti, E. Corradini, M. Sabelli, S. Canali, A. Pietrangelo, Gluconeogenic signals regulate iron homeostasis via hepcidin in mice, Gastroenterology 146 (2014) 1060–1069.
[5] S. Gordon, G. Akopyan, H. Garban, B. Bonavida, Transcription factor YY: structure, function, and therapeutic implications in cancer biology, Oncogene 25 (2006) 1125–1142.
[6] Z. Deng, P. Cao, M.M. Wan, G. Sui, Yin Yang 1: a multifaceted protein beyond a transcription factor, Transcription 1 (2010) 81–84.
[7] Y. Lu, X. Xiong, X. Wang, Z. Zhang, J. Li, G. Shi, J. Yang, H. Zhang, G. Ning, X. Li, Ying Yang 1 promotes hepatic gluconeogenesis through upregulation of glucocorticoid receptor, Diabetes 62 (2013) 1064–1073.
[8] Y. Lu, Z. Ma, Z. Zhang, X. Xiong, X. Wang, H. Zhang, G. Shi, X. Xia, G. Ning, X. Li, Ying Yang 1 promotes hepatic steatosis through repression of farnesoid X receptor in obese mice, Gut 63 (2014) 170–178.
[9] F. Mauxion, C.Y. Chen, B. Séraphin, A.B. Shyu, BTG/TOB factors impact deadenylases, Trends. Biochem. Sci. 34 (2009) 640–647.
[10] G.S. Winkler, The mammalian anti-proliferative BTG/Tob protein family, J. Cell. Physiol. 222 (2010) 66–72.
[11] S.L. Hwang, O. Kwon, S.J. Lee, Y.D. Kim, J.H. Choi, B-cell translocation gene-2 increases hepatic gluconeogenesis via induction of CREB, Biochem. Biophys. Res. Commun. 427 (2012) 801–805.
[12] S.L. Hwang, O. Kwon, S.G. Kim, I.K. Lee, Y.D. Kim, B-cell translocation gene 2 positively regulates GLP-1-stimulated insulin secretion via induction of PDX-1 in pancreatic β-cells, Exp. Mol. Med. 45 (2013) e25.
[13] Y.D. Kim, S.G. Kim, S.L. Hwang, H.S. Choi, J.H. Bae, D.K. Song, S.S. Im, B-cell translocation gene 2 regulates hepatic glucose homeostasis via induction of orphan nuclear receptor Nur77 in diabetic mouse model, Diabetes 63 (2014) 1870–1880.
[14] D.K. Kim, J.H. Jeong, J.M. Lee, K.S. Kim, S.H. Park, Y.D. Kim, M. Koh, M. Shin, Y.S. Jung, H.S. Kim, T.H. Lee, B.C. Oh, J.I. Kim, H.T. Park, W.I. Jeong, C.H. Lee, S.B. Park, J.J. Min, S.I. Jung, S.Y. Choi, H.E. Choy, H.S. Choi, Inverse agonist of estrogen-related receptor γ controls Salmonella typhimurium infection by modulating host iron homeostasis, Nat. Med. 20 (2014) 419–424.
[15] Y.D. Kim, K.G. Park, Y.S. Lee, Y.Y. Park, D.K. Kim, B. Nedumaran, W.G. Jang, W.J. Cho, J. Ha, I.K. Lee, C.H. Lee, H.S. Choi, Metformin inhibits hepatic gluconeogenesis through AMP-activated protein kinase-dependent regulation of the orphan nuclear receptor SHP, Diabetes 57 (2008) 306–314.
[16] J.C. Deschemic, M. Foretz, B. Viollet, S. Vaulont, Hepatic peroxisome proliferator-activated receptor γ coactivator 1α and hepcidin are coregulated in fasted/refed states in mice, Clin. Chem. 58 (2012) 1487–1488.
[17] J.S. Troutt, M. Rudling, L. Persson, L. Ståhle, B. Angelin, A.M. Butterfield, A.E. Schade, G. Cao, R.J. Konrad, Circulating human hepcidin-25 concentrations display a diurnal rhythm, increase with prolonged fasting, and are reduced by growth hormone administration, Clin. Chem. 58 (2012) 1225–1232.
[18] J.T. Cunningham, J.T. Rodgers, D.H. Arlow, F. Vazquez, V.K. Mootha, P. Puigserver, mTOR controls mitochondrial oxidative function through a YY1-PGC-1alpha transcriptional complex, Nature 450 (2007) 736–740.