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Lessons from new mouse models of glycogen storage disease type 1a  
in relation to the time course and organ specificity of the disease

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## Summary

1  
2 Patients with glycogen storage diseases type 1 (GSD1) suffer from life-threatening  
3  
4 hypoglycaemia, when left untreated. Despite an intensive dietary treatment, patients develop  
5  
6 severe complications, such as liver tumors and renal failure, with aging. Until now, the animal  
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8 models available for studying the GSD1 did not survive after weaning. To gain further  
9  
10 insights into the molecular mechanisms of the disease and to evaluate potential treatment  
11  
12 strategies, we have recently developed novel mouse models in which the catalytic subunit of  
13  
14 glucose-6 phosphatase (*G6pc*) is deleted in each glucose-producing organ specifically. For  
15  
16 that, B6.*G6pc*<sup>ex3lox/ex3lox</sup> mice were crossed with transgenic mice expressing a recombinase  
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18 under the control of the serum albumin, the kidney androgen protein or the villin promoter, in  
19  
20 order to obtain liver, kidney or intestine *G6pc*<sup>-/-</sup> mice, respectively.  
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26 As opposed to total *G6pc* knockout mice, tissue-specific *G6pc* deficiency allows mice  
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28 to maintain their blood glucose by inducing glucose production in the other gluconeogenic  
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30 organs. Even though it is considered that glucose is produced mainly by the liver, liver *G6pc*<sup>-/-</sup>  
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32 mice are perfectly viable and exhibit the same hepatic pathological features as GSD1 patients,  
33  
34 including the late development of hepatocellular adenomas and carcinomas. Interestingly,  
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36 renal *G6pc*<sup>-/-</sup> mice developed renal symptoms similar to the early human GSD1 nephropathy.  
37  
38 This includes glycogen overload that leads to nephromegaly and morphological and  
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40 functional alterations in the kidneys. Thus, our data suggest that renal G6Pase deficiency *per*  
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42 *se* is sufficient to induce the renal pathology of GSD1.  
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48 Therefore, these new mouse models should allow us to improve the strategies of  
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50 treatment on both nutritional and pharmacological points of view.  
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56 **Synopsis:** Mice with tissue-specific G6Pase deletion allowed the reproduction of the hepatic  
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58 and renal pathologies of GSD1a that occur with aging.  
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1 **Compliance with Ethics Guidelines:**  
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6 **Animal rights**  
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8 All of the procedures were performed in accordance with the principles and guidelines  
9 established by the European Convention for the Protection of Laboratory Animals. The  
10 animal care committee of University Lyon 1 approved all of the experiments.  
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18 **Details of the contributions of individual authors**  
19

20 Fabienne Rajas is the PI of this study. Julie Clar is a PhD student who characterized the renal  
21 GSD1a mouse model. Amandine Gautier-Stein is a researcher who characterized glucose  
22 transport by the intestine. Gilles Mithieux is the director of the lab.  
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30 **Competing Interests**  
31

32 Fabienne Rajas, Julie Clar, Amandine Gautier-Stein and Gilles Mithieux declare that they  
33 have no conflict of interest.  
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5 Glycogen storage disease type 1 (GSD1) is a rare metabolic disorder characterized by  
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7 the absence of endogenous glucose production, leading to severe hypoglycemia following a  
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9 short fast (Chou et al. 2010; Froissart et al. 2011). This is caused by a deficiency of glucose-6  
10  
11 phosphatase (G6Pase), which is an enzyme complex involving the glucose-6 phosphate  
12  
13 translocase subunit (G6PT, encoded by *SLC37A4*) and the G6Pase catalytic subunit, encoded  
14  
15 by *G6PCI* (Soty et al. 2012) (Figure 1). Patients with GSD type 1a (GSD1a) represent 80%  
16  
17 of all GSD1 cases and have a G6PC deficiency, whereas patients with GSD type 1b (GSD1b)  
18  
19 have a G6PT defect. While G6PT is ubiquitously expressed, G6PC is expressed only in the  
20  
21 liver, kidneys and intestine (Rajas et al. 1999; Mithieux et al. 2004b; Rajas et al.  
22  
23 2007a)(Mithieux et al. 2004b; Rajas et al. 2007b). Both GSD1a and GSD1b patients show  
24  
25 broadly similar symptoms, including severe hypoglycemia in the post-absorptive state,  
26  
27 hyperlipidemia, hypercholesterolemia, hyperuricemia and lactic acidemia. G6Pase deficiency  
28  
29 leads to the accumulation of glycogen and triglycerides in the liver and kidneys. This results  
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31 in marked hepatomegaly, nephromegaly and hepatic steatosis. In addition, patients with  
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33 GSD1b present severe infectious complications, due to neutropenia and neutrophil and  
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35 monocyte functional defects.

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Until today, there is no cure for GSD1. In the past, many patients with GSD1 did not survive infancy and childhood. Since the eighties, life expectancy of patients with GSD1 has been considerably improved by stringent dietary treatment (Rake et al. 2002a; Heller et al. 2008). Frequent meals combined with uncooked cornstarch (during the day- and/or night-time) or gastric drip-feeding allow patients with GSD1 to avoid hypoglycemia and lactic acidemia. Despite the intensive dietary treatment, hepatic, renal and intestinal complications arise with aging (Di Rocco et al. 2008; Reddy et al. 2009). Thus a large proportion of patients

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older than 20 years show hepatic tumors and/or progressive chronic renal disease, which are the leading causes of morbidity in GSD1 patients with aging. The first hepatic tumors generally appear during adolescence and are mainly classified as hepatocellular adenomas (HCA), which can transform into hepatocellular carcinomas (HCC) (Rake et al. 2002b; Franco et al. 2005; Wang et al. 2011; Calderaro et al. 2013). Tumor resection or liver transplantation are recommended if the tumors are associated with serious compression or hemorrhage, or show signs of malignant transformation into HCC (Rake et al. 2002a; Reddy et al. 2009). The first symptoms of renal disease are hyperperfusion and hyperfiltration and generally appear from childhood (Martens et al. 2009). Almost all adult patients show albuminuria and more than 50% present proteinuria (Martens et al. 2009). Finally, renal disease can slowly progress into renal failure that requires renal dialysis or transplantation (Rake et al. 2002a).

In order to investigate the onset of long-term pathologies developed by patients with GSD1a and to evaluate potential treatment strategies, we recently developed new mouse models of GSD1a that exclusively target G6PC deletion in the liver, kidneys or intestine (Mutel et al. 2011a; Penhoat et al. 2011; Clar et al. 2014). It is noteworthy that these tissue-specific G6PC knockout mice have normal life expectancy, whereas total G6PC knockout mouse and canine models die rapidly after weaning in the absence of intensive dietary therapy. In this review, we propose an overview of these new murine GSD1a models with tissue-specific *G6pc* deletions.

### **The canine and mouse models of total *G6pc* deficiency**

Until now, the understanding of the biochemical bases of GSD1a and the evaluation of gene therapy approaches to correct *G6pc* deficiency were performed in two animal models of GSD1a. Both of these animal models are physiologically similar to humans in regard of the

1 glucose-6 phosphate metabolism. After the isolation of the *G6pc* gene, Dr. Janice Chou  
2 created the *G6pc* knockout mouse in 1993 (Lei et al. 1996). These mice present low birth  
3 weight, develop quickly severe and unremitting hypoglycemia quickly and gradually display  
4 pronounced increases in serum cholesterol and triglyceride levels (Table 1). However, they do  
5 not typically manifest lactic acidemia. Moreover, only 60% of mice treated with glucose  
6 injections, glucose-fortified water and food supplementation survive through weaning and  
7 have a life expectancy of about six months. **Although all *G6pc* knockout mice developed  
8 hepatomegaly and steatosis (Table 1), no hepatic tumors have been reported, even in mice as  
9 old as 6 months (Salganik et al. 2009). These mice exhibited nephromegaly as well (Table 1).  
10 Nevertheless, the renal pathology has been characterized only in an early stage (in 6-week-old  
11 KO mice) (Yiu et al. 2008).** A GSD1a canine model (Maltese breed) carrying a natural *G6pc*  
12 mutation was identified and used to characterize the disease as well (Kishnani et al. 1997).  
13 The Maltese breed is small in size and exhibits low survival rate of newborns. A second  
14 canine model was obtained by crossbreeding a carrier Maltese with beagles (Kishnani et al.  
15 2001). Both canine models manifest all of the typical symptoms of the human disorder,  
16 including hyperlactacidemia, but they did not prove useful for studying the long-term  
17 complications of the GSD1a. Indeed, the dietary therapies used to maintain GSD1a animals  
18 viable have not yet been sufficiently refined to prevent premature death of these animals from  
19 hypoglycemia.

### **Rationale underlying the generation of organ-specific *G6pc* deficient mice.**

20 The maintenance of blood glucose levels within a narrow range (about 5.5 mmol/L) is  
21 a critical physiological function. Although the liver is the main glucose-producing organ in  
22 the post-absorptive state *via* glycogenolysis, renal and intestinal glucose productions play a  
23 major role in maintaining normoglycemia during long fasting periods (Ekberg et al. 1999;  
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1 Gerich et al. 2001; Croset et al. 2001; Mithieux et al. 2004a). We hypothesized that **total**  
2 inactivation of *G6pc* in only one gluconeogenic tissue would not be lethal due to the  
3  
4 compensatory **induction of glucose** production by the two non-targeted tissues. To delete the  
5  
6 G6Pase activity, we targeted the excision of *G6pc* exon 3, by using B6.G6pc<sup>ex3lox/ex3lox</sup> mice.  
7  
8 We crossed B6.G6pc<sup>ex3lox/ex3lox</sup> mice with mice expressing a CRE recombinase under a tissue-  
9  
10 specific promoter to target the *G6pc* deletion in the liver, kidney or intestine specifically. As  
11  
12 endogenous glucose production, especially in the liver, is critical during the neonatal period  
13  
14 because of the low content of glucose in milk (Girard et al. 1992; Chatelain et al. 1998), we  
15  
16 chose to induce *G6pc* deletion in adult mice by using a CRE<sup>ERT2</sup> inducible by tamoxifen. The  
17  
18 CRE<sup>ERT2</sup> is a recombinant CRE, fused to a mutated ligand-binding protein of the estrogen,  
19  
20 resulting in a tamoxifen-dependent CRE. In order to induce *G6pc* deletion, adult mice are  
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22 treated daily with 1 mg of tamoxifen for 5 consecutive days.  
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### 31 **Liver *G6pc* knockout mice.**

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34 Hepatic *G6pc* deletion was targeted by crossing B6.G6pc<sup>ex3lox/ex3lox</sup> mice with  
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36 B6.SA<sup>CREERT2/w</sup> mice, which expressed the CRE<sup>ERT2</sup> under the liver-specific serum albumin  
37  
38 promoter. Rapidly (10 days) after tamoxifen treatment, hepatic G6Pase activity was  
39  
40 undetectable in liver-specific *G6pc* knockout (L.G6pc<sup>-/-</sup>) mice (Mutel et al. 2011a). As  
41  
42 expected, L.G6pc<sup>-/-</sup> mice had normal life expectancy and showed normal blood glucose  
43  
44 during fed state (Table 1). Interestingly, L.G6pc<sup>-/-</sup> mice did not exhibit marked hypoglycemia  
45  
46 during long fasting periods thanks to the induction of extrahepatic glucose production (Mutel  
47  
48 et al. 2011b; Penhoat et al. 2014). The livers of L.G6pc<sup>-/-</sup> mice were rapidly enlarged, due to  
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50 the accumulation of glycogen and triglycerides (Table 1). In parallel, there was a rapid  
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52 increase in plasma triglyceride, cholesterol, uric acid, and lactic acid levels (Table 1).  
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58 However, these plasmatic parameters (except for cholesterol) improved after 6 months, as  
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observed in total *G6pc* knockout mice, which survive weaning thanks to a diet therapy (Salganik et al. 2009). We suggested that a satisfying blood glucose control could explain this amelioration. This is also in line with the observation that metabolic control is easier in adult patients with GSD1a, than during infancy or childhood.

Contrary to plasmatic parameters, glycogen storages were still elevated after 18 months of *G6pc* deletion. It is noteworthy that the accumulation of triglycerides increased with time. This could be linked to the late development of hepatocellular adenomas observed in L.*G6pc*<sup>-/-</sup> mice fed a standard diet. The first millimetric nodules were detected by magnetic resonance imaging after 9 months of *G6pc* deletion in about 20% of L.*G6pc*<sup>-/-</sup> mice. Most of them developed multiple nodules of about 1 to 10 mm in diameter after 18 months of *G6pc* deletion. Most of these tumors were HCA, but some L.*G6pc*<sup>-/-</sup> livers (20%) presented dysplasia as well (Mutel et al. 2011a). **In addition, about 5-10% of L.*G6pc*<sup>-/-</sup> mice fed a standard diet developed HCC (unpublished data).** It is important to note that the development of tumors appeared rather late, while liver steatosis tended to worsen. Moreover, our recent data show that the development of hepatic tumors in L.*G6pc*<sup>-/-</sup> mice is enhanced by a high fat enriched diet (to be published). To propose therapeutic strategies or to update the dietary advices, it is now important to determine the molecular mechanisms involved in tumor development and to understand how is tumorigenesis influenced by the hepatic metabolism. Thus L.*G6pc*<sup>-/-</sup> mice would be a unique model that could allow us to answer these questions and to study the effect of diet on the development of HCA and HCC.

### **Kidney *G6pc* knockout mice.**

Renal *G6pc* deletion was targeted by crossing B6.*G6pc*<sup>ex3lox/ex3lox</sup> mice with B6.Kap<sup>CREERT2/w</sup> mice, which expressed the CRE<sup>ERT2</sup> under the kidney androgen-regulated protein (Kap) promoter (Clar et al. 2014). This resulted in a partial lost (50% of inhibition) of

1 G6Pase activity after tamoxifen treatment of adult male mice. Immunohistological  
2 observations of K-G6pc<sup>-/-</sup> kidneys revealed a lower and heterogeneous staining of G6PC in  
3  
4 the cortex with the presence of only a few G6PC-positive cells identified in the proximal  
5  
6 convoluted tubules. Female B6.G6pc<sup>ex3lox/ex3lox</sup>.Kap<sup>CREERT2/w</sup> mice have to be treated with both  
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8 testosterone and tamoxifen to induce *G6pc* deletion. As expected, K-G6pc<sup>-/-</sup> mice did not  
9  
10 suffer from hypoglycemia and did not require diet treatment. This is partially due to the  
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12 residual renal G6Pase activity and to a compensatory induction of hepatic G6Pase activity  
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14 (Clar et al. 2014). It is noteworthy that K-G6pc<sup>-/-</sup> mice showed an early stage nephropathy i.e.  
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16 microalbuminuria and a partial electrolyte imbalance, after 6 months of *G6pc* deletion (Table  
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18 1) (Clar et al. 2014). However, they did not develop proteinuria, kidney fibrosis and  
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20 nephrolithiasis. On a molecular level, the accumulation of glucose-6 phosphate activated the  
21  
22 *de novo* lipogenesis pathway. The slight accumulation of triglycerides observed in the K-  
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24 G6pc<sup>-/-</sup> kidney by red Sudan staining was sufficient to activate the renin-angiotensin system,  
25  
26 which was associated with an increased expression of the transforming growth factor  $\beta$ 1  
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28 (TGF $\beta$ 1). This led to partial epithelial-mesenchymal transition (EMT)-like changes,  
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30 highlighted by a decrease of epithelial marker expression (e.g. E-cadherin, tight junction ZO-  
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32 1) and an overexpression of the mesenchymal marker fibronectin. Podocyte injury  
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34 characterized by a decrease in podocin, synaptopodin and podocalyxin expression was  
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36 observed in K-G6pc<sup>-/-</sup> kidneys. However, no modifications in the thickness of basement  
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38 membrane or glomerulosclerosis were observed, suggesting that K-G6pc<sup>-/-</sup> kidneys showed an  
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40 early stage of EMT only after 6 months of *G6pc* deletion (Clar et al. 2014).  
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51 The characterization of renal K-G6pc<sup>-/-</sup> metabolism allowed us to highlight similarities  
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53 of molecular pathways involved in the development of EMT in both GSD1 and diabetes  
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55 (Mundy and Lee 2002; Rajas et al. 2013). Indeed, the accumulation of lipids or lipid  
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57 derivatives leads to the activation of renin- angiotensin system and subsequently of the  
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1 TGFβ1 pathway. The presence of lipid deposits in the renal cortex of two GSD1 patients with  
2 proteinuria was reported in only one study (Obara et al. 1993). As opposed to diabetic  
3 nephropathy, renal lipid accumulation in K-G6pc<sup>-/-</sup> mice did not seem to be mediated by the  
4 transcriptional factors SREBP1 (sterol regulatory element-binding protein), but rather by  
5 ChREBP (carbohydrate-responsive element binding protein) (Clar et al. 2014). Compared to  
6 the liver, little is known about lipid metabolism and lipid deposits in the kidney. The K-G6pc<sup>-/-</sup>  
7 mouse model is therefore a unique tool that can be used to decipher the role of lipids,  
8 compared to glycogen accumulation, in the development of nephropathy.  
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### 22 **Intestinal *G6pc* knockout mice.**

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24 Until now, intestinal symptoms were often underestimated in GSD1a patients, when  
25 compared to GSD1b in whom they appear more serious (Visser et al. 2002). However, some  
26 studies reported that GSD1a patients might also suffer from intermittent diarrhea due to  
27 entero-proctitis (Fine et al. 1969; Milla et al. 1978; Rake et al. 2002b). An abnormal  
28 accumulation of glycogen has also been reported in the intestine of GSD1a patients (Field et  
29 al. 1965).  
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39 Intestinal *G6pc* deletion was targeted by crossing B6.G6pc<sup>ex3lox/ex3lox</sup> mice with  
40 B6.Villin<sup>CREERT2/w</sup> mice, which express the CRE<sup>ERT2</sup> under the Villin promoter (Penhoat et al.  
41 2011). The specific deletion of *G6pc* in the intestine of I-G6pc<sup>-/-</sup> mice was persistent for more  
42 than one year. During the first year after *G6pc* gene deletion, I-G6pc<sup>-/-</sup> mice exhibited a  
43 growth rate similar to that of wild-type mice. Meanwhile, no diarrhea or abnormal  
44 consistency of the stools was observed in any I-G6pc<sup>-/-</sup> mice, compared to wild-type mice.  
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1 influence intestine metabolism. Hence, we cannot exclude that some adverse effects could  
2 appear with time according to the diet composition. This hypothesis will be assessed in the I-  
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4  $G6pc^{-/-}$  mice in the near future.  
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7 For example, the ingestion of too much sugar, including uncooked starched, in patients  
8 with GSD1 could lead to bacterial proliferation. A bacterial overgrowth has been documented  
9 in a GSD1b patient by using a hydrogen breath test (Santer et al. 2003). In the latter study, the  
10 authors suggested that the absence of a functional G6Pase might induce carbohydrate  
11 malabsorption by the intestine and promote bacterial overgrowth. Indeed, G6Pase has been  
12 suggested to be involved in intestinal transepithelial glucose transport. The transepithelial  
13 glucose transport in the intestine is thought to mainly involve the sodium-dependent glucose  
14 cotransporter SGLT1 at the apical membrane and the glucose transporter GLUT2 at the basal  
15 membrane. However, glucose absorption, challenged by an oral glucose test, is similar in  
16  $Glut2^{-/-}$  mice and wild-type mice, suggesting the existence of another intestinal glucose  
17 transport pathway (Stümpel et al. 2001). The absorption of glucose measured in isolated  
18 perfused intestine and liver of  $Glut2^{-/-}$  mice is inhibited by S4048, a specific inhibitor of  
19 G6PT. In the absence of GLUT2, a step of phosphorylation (by hexokinase) and hydrolysis  
20 (by G6Pase) seems thus needed for glucose transport through the intestine. This would be  
21 consistent with a high hexokinase activity in the intestine, 10-fold greater than the reported  
22 maximal flux of glucose through glycolysis (Newsholme and Carrié 1994). Finally, the gut  
23 microbiota fermentation is tightly coupled to intestinal metabolism (De Vadder et al. 2014). I-  
24  $G6pc^{-/-}$  mice are thus a unique model that can be used to study the precise role of intestinal  
25 glucose absorption and metabolism in the etiology of intestinal symptoms of GSD1 patients.  
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## Lessons from the tissue-specific *G6pc* knockout mouse models

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2 The usefulness of these new animal models of GSD1a is based primarily on the fact  
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4 that these animals are viable. They permitted the characterization of the development of  
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6 hepatic tumors and nephropathy during several months. These results suggest that the hepatic  
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8 or renal *G6pc* deficiency *per se* is sufficient to induce independently the development of  
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10 hepatic or renal pathology, respectively. This highlighted the fact that the development of  
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12 nephropathy is independent of the liver-derived metabolic changes. This was an important  
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14 finding in the context of the pathophysiological concept of the GSD1 disease as a whole, but  
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16 also in terms of transplantation or gene therapy. Kidney transplantation is performed in the  
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18 case of severe renal failure, but this does not correct hypoglycemia (Emmett and Narins  
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20 1978). In contrast, liver transplantation allows a correction of the glycemia and all liver-  
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22 related biochemical abnormalities, but any beneficial effects on the kidney remain to be  
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24 proven (Faivre et al. 1999; Labrune 2002; Davis and Weinstein 2008; Reddy et al. 2009).  
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26 This finding suggests that clinicians should discuss the long-term benefits and possible  
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28 convenience of a double kidney/liver transplantation for GSD1a patients, who have developed  
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30 multiple liver HCAs or HCCs or renal failure.  
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39 Concerning gene therapy, it was already shown that the rescue of G6Pase activity in  
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41 the liver by using an AAV8 recombinant vector allowed total *G6pc* KO mice to maintain  
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43 normal blood glucose levels, but it did not prevent the development of the nephropathy (Yiu  
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45 et al. 2010; Luo et al. 2011). As AAV1 or AAV2/9 vectors are able to transduce both the liver  
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47 and the kidney, these vectors seem to be a better choice for GSD1a gene therapy (Ghosh et al.  
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49 2006; Luo et al. 2011). Thus, the hepatic and renal  $G6pc^{-/-}$  mouse models will be useful to test  
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51 the efficiency and the safety of gene therapy. In addition,  $L.G6pc^{-/-}$  mice could be used to test  
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53 drugs targeting the development of hepatic tumors. Interestingly, the molecular mechanisms  
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55 involved in GSD1a nephropathy are very similar to those of diabetic patients (Mundy and Lee  
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2002; Rajas et al. 2013). Therefore, K.G6pc<sup>-/-</sup> mice could be a useful tool in future studies of the pharmacological treatment of EMT and/or kidney failure developed by patients with either GSD1 or diabetes.

Finally, these mice should allow us to improve the strategies of treatment on a nutritional point of view. Indeed, preliminary results showed that diet could greatly influence the development of hepatic tumors in L.G6pc<sup>-/-</sup> mice (to be published). Moreover, until now, dietary guidelines were only based on biochemical knowledge and vary greatly between pediatricians. For example, some clinicians prohibit entirely the consumption of fruits, juice, dairy products and sweets. Others only limit the consumption of these products, in order to restrain the intake of fructose, lactose and sucrose. To provide scientific evidence, L.G6pc<sup>-/-</sup> and K.G6pc<sup>-/-</sup> mice will be useful to analyze the effect of diet on the development of hepatic tumors and nephropathy, respectively. Meanwhile, it is important to remind teenagers and adults to follow strictly their diet, even if they already exhibit a good metabolic control, in order to delay or avoid the development of the GSD1 complications.

#### **Legend of figure:**

**Figure 1: Schematic representation of the glucose-6 phosphatase.** This enzyme is composed of two subunits localized in the membrane of the endoplasmic reticulum: the transport subunit (G6PT) and the catalytic subunit (G6PC). The G6PT subunit is ubiquitously expressed, whereas the G6PC is expressed only in the liver, kidneys and intestine. The mutations of G6PC are responsible for GSD type 1a and the mutations of G6PT are responsible for GSD type 1b. Images were made with Servier Medical Art illustrations.

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**References:**

12  
13  
14  
15  
16  
17  
18  
Calderaro J, Labrune P, Morcrette G, et al. (2013) Molecular characterization of hepatocellular adenomas developed in patients with glycogen storage disease type I. *J Hepatol* 58:350–357. doi: 10.1016/j.jhep.2012.09.030

19  
20  
21  
22  
23  
24  
25  
26  
Chatelain F, Pégorier JP, Minassian C, et al. (1998) Development and regulation of glucose-6-phosphatase gene expression in rat liver, intestine, and kidney: in vivo and in vitro studies in cultured fetal hepatocytes. *Diabetes* 47:882–889.

27  
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30  
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57  
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60  
61  
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63  
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65  
Chou JY, Jun HS, Mansfield BC (2010) Glycogen storage disease type I and G6Pase- $\beta$  deficiency: etiology and therapy. *Nat Rev Endocrinol* 6:676–688. doi: 10.1038/nrendo.2010.189

Clar J, Gri B, Calderaro J, et al. (2014) Targeted deletion of kidney glucose-6 phosphatase leads to nephropathy. *Kidney Int.* doi: 10.1038/ki.2014.102

Croset M, Rajas F, Zitoun C, et al. (2001) Rat small intestine is an insulin-sensitive gluconeogenic organ. *Diabetes* 50:740–746.

Davis MK, Weinstein DA (2008) Liver transplantation in children with glycogen storage disease: controversies and evaluation of the risk/benefit of this procedure. *Pediatr Transplant* 12:137–145. doi: 10.1111/j.1399-3046.2007.00803.x

De Vadder F, Kovatcheva-Datchary P, Goncalves D, et al. (2014) Microbiota-generated metabolites promote metabolic benefits via gut-brain neural circuits. *Cell* 156:84–96. doi: 10.1016/j.cell.2013.12.016

Di Rocco M, Calevo MG, Taro' M, et al. (2008) Hepatocellular adenoma and metabolic

1 balance in patients with type Ia glycogen storage disease. *Mol Genet Metab* 93:398–402. doi:  
2 10.1016/j.ymgme.2007.10.134  
3

4 Ekberg K, Landau BR, Wajngot A, et al. (1999) Contributions by kidney and liver to  
5 glucose production in the postabsorptive state and after 60 h of fasting. *Diabetes* 48:292–298.  
6

7 Emmett M, Narins RG (1978) Renal transplantation in type 1 glycogenosis. Failure to  
8 improve glucose metabolism. *JAMA J Am Med Assoc* 239:1642–1644.  
9

10 Faivre L, Houssin D, Valayer J, et al. (1999) Long-term outcome of liver transplantation  
11 in patients with glycogen storage disease type Ia. *J Inherit Metab Dis* 22:723–732.  
12

13 Field JB, Epstein S, Egan T (1965) Studies in glycogen storage diseases. I. Intestinal  
14 glucose-6 phosphatase activity in patients with Von Gierke's disease and their parents. *J Clin*  
15 *Invest* 44:1240–1247. doi: 10.1172/JCI105230  
16

17 Fine RN, Kogut MD, Donnell GN (1969) Intestinal absorption in type I glycogen storage  
18 disease. *J Pediatr* 75:632–635.  
19

20 Franco LM, Krishnamurthy V, Bali D, et al. (2005) Hepatocellular carcinoma in  
21 glycogen storage disease type Ia: a case series. *J Inherit Metab Dis* 28:153–162. doi:  
22 10.1007/s10545-005-7500-2  
23

24 Froissart R, Piraud M, Boudjemline AM, et al. (2011) Glucose-6-phosphatase deficiency.  
25 *Orphanet J Rare Dis* 6:27. doi: 10.1186/1750-1172-6-27  
26

27 Gerich JE, Meyer C, Woerle HJ, Stumvoll M (2001) Renal gluconeogenesis: its  
28 importance in human glucose homeostasis. *Diabetes Care* 24:382–391.  
29

30 Ghosh A, Allamarvdasht M, Pan C-J, et al. (2006) Long-term correction of murine  
31 glycogen storage disease type Ia by recombinant adeno-associated virus-1-mediated gene  
32 transfer. *Gene Ther* 13:321–329. doi: 10.1038/sj.gt.3302650  
33

34 Girard J, Ferré P, Pégrier JP, Duée PH (1992) Adaptations of glucose and fatty acid  
35 metabolism during perinatal period and suckling-weaning transition. *Physiol Rev* 72:507–  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
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48  
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56  
57  
58  
59  
60  
61  
62  
63  
64  
65



562.

1  
2 Heller S, Worona L, Consuelo A (2008) Nutritional therapy for glycogen storage  
3 diseases. *J Pediatr Gastroenterol Nutr* 47 Suppl 1:S15–21. doi:  
4  
5 10.1097/MPG.0b013e3181818ea5  
6  
7

8  
9 Kim SY, Weinstein DA, Starost MF, et al. (2008) Necrotic foci, elevated chemokines and  
10 infiltrating neutrophils in the liver of glycogen storage disease type Ia. *J Hepatol* 48:479–485.  
11  
12 doi: 10.1016/j.jhep.2007.11.014  
13  
14

15  
16 Kishnani PS, Bao Y, Wu JY, et al. (1997) Isolation and nucleotide sequence of canine  
17 glucose-6-phosphatase mRNA: identification of mutation in puppies with glycogen storage  
18  
19 disease type Ia. *Biochem Mol Med* 61:168–177.  
20  
21  
22

23  
24 Kishnani PS, Faulkner E, VanCamp S, et al. (2001) Canine model and genomic structural  
25 organization of glycogen storage disease type Ia (GSD Ia). *Vet Pathol* 38:83–91.  
26  
27

28  
29 Labrune P (2002) Glycogen storage disease type I: indications for liver and/or kidney  
30 transplantation. *Eur J Pediatr* 161 Suppl 1:S53–55. doi: 10.1007/s00431-002-1004-y  
31  
32

33  
34 Lei KJ, Chen H, Pan CJ, et al. (1996) Glucose-6-phosphatase dependent substrate  
35 transport in the glycogen storage disease type-Ia mouse. *Nat Genet* 13:203–209. doi:  
36  
37 10.1038/ng0696-203  
38  
39

40  
41 Luo X, Hall G, Li S, et al. (2011) Hepatorenal correction in murine glycogen storage  
42 disease type I with a double-stranded adeno-associated virus vector. *Mol Ther J Am Soc Gene*  
43  
44 *Ther* 19:1961–1970. doi: 10.1038/mt.2011.126  
45  
46

47  
48 Martens DHJ, Rake JP, Navis G, et al. (2009) Renal function in glycogen storage disease  
49 type I, natural course, and renopreservative effects of ACE inhibition. *Clin J Am Soc Nephrol*  
50  
51 *CJASN* 4:1741–1746. doi: 10.2215/CJN.00050109  
52  
53

54  
55 Milla PJ, Atherton DA, Leonard JV, et al. (1978) Disordered intestinal function in  
56  
57 glycogen storage disease. *J Inherit Metab Dis* 1:155–157.  
58  
59  
60  
61  
62  
63  
64  
65

1 Mithieux G, Bady I, Gautier A, et al. (2004a) Induction of control genes in intestinal  
2 gluconeogenesis is sequential during fasting and maximal in diabetes. *Am J Physiol*  
3  
4 *Endocrinol Metab* 286:E370–375. doi: 10.1152/ajpendo.00299.2003  
5  
6

7 Mithieux G, Rajas F, Gautier-Stein A (2004b) A novel role for glucose 6-phosphatase in  
8  
9 the small intestine in the control of glucose homeostasis. *J Biol Chem* 279:44231–44234. doi:  
10  
11 10.1074/jbc.R400011200  
12  
13

14 Mundy HR, Lee PJ (2002) Glycogenesis type I and diabetes mellitus: a common  
15  
16 mechanism for renal dysfunction? *Med Hypotheses* 59:110–114.  
17  
18

19 Mutel E, Abdul-Wahed A, Ramamonjisoa N, et al. (2011a) Targeted deletion of liver  
20  
21 glucose-6 phosphatase mimics glycogen storage disease type 1a including development of  
22  
23 multiple adenomas. *J Hepatol* 54:529–537. doi: 10.1016/j.jhep.2010.08.014  
24  
25

26 Mutel E, Gautier-Stein A, Abdul-Wahed A, et al. (2011b) Control of blood glucose in the  
27  
28 absence of hepatic glucose production during prolonged fasting in mice: induction of renal  
29  
30 and intestinal gluconeogenesis by glucagon. *Diabetes* 60:3121–3131. doi: 10.2337/db11-0571  
31  
32  
33

34 Newsholme EA, Carrié AL (1994) Quantitative aspects of glucose and glutamine  
35  
36 metabolism by intestinal cells. *Gut* 35:S13–17.  
37  
38

39 Obara K, Saito T, Sato H, et al. (1993) Renal histology in two adult patients with type I  
40  
41 glycogen storage disease. *Clin Nephrol* 39:59–64.  
42  
43

44 Penhoat A, Fayard L, Stefanutti A, et al. (2014) Intestinal gluconeogenesis is crucial to  
45  
46 maintain a physiological fasting glycemia in the absence of hepatic glucose production in  
47  
48 mice. *Metabolism* 63:104–111. doi: 10.1016/j.metabol.2013.09.005  
49  
50

51 Penhoat A, Mutel E, Amigo-Correig M, et al. (2011) Protein-induced satiety is abolished  
52  
53 in the absence of intestinal gluconeogenesis. *Physiol Behav* 105:89–93. doi:  
54  
55 10.1016/j.physbeh.2011.03.012  
56  
57

58 Rajas F, Bruni N, Montano S, et al. (1999) The glucose-6 phosphatase gene is expressed  
59  
60  
61  
62  
63  
64  
65

1  
2 in human and rat small intestine: regulation of expression in fasted and diabetic rats.  
3 Gastroenterology 117:132–139.

4  
5 Rajas F, Jourdan-Pineau H, Stefanutti A, et al. (2007a) Immunocytochemical localization  
6  
7 of glucose 6-phosphatase and cytosolic phosphoenolpyruvate carboxykinase in gluconeogenic  
8  
9 tissues reveals unsuspected metabolic zonation. Histochem Cell Biol 127:555–565. doi:  
10  
11 10.1007/s00418-006-0263-5  
12

13  
14 Rajas F, Jourdan-Pineau H, Stefanutti A, et al. (2007b) Immunocytochemical localization  
15  
16 of glucose 6-phosphatase and cytosolic phosphoenolpyruvate carboxykinase in gluconeogenic  
17  
18 tissues reveals unsuspected metabolic zonation. Histochem Cell Biol 127:555–565. doi:  
19  
20 10.1007/s00418-006-0263-5  
21  
22

23  
24 Rajas F, Labrune P, Mithieux G (2013) Glycogen storage disease type 1 and diabetes:  
25  
26 Learning by comparing and contrasting the two disorders. Diabetes Metab. doi:  
27  
28 10.1016/j.diabet.2013.03.002  
29  
30

31  
32 Rake JP, Visser G, Labrune P, et al. (2002a) Guidelines for management of glycogen  
33  
34 storage disease type I - European Study on Glycogen Storage Disease Type I (ESGSD I). Eur  
35  
36 J Pediatr 161 Suppl 1:S112–119. doi: 10.1007/s00431-002-1016-7  
37

38  
39 Rake JP, Visser G, Labrune P, et al. (2002b) Glycogen storage disease type I: diagnosis,  
40  
41 management, clinical course and outcome. Results of the European Study on Glycogen  
42  
43 Storage Disease Type I (ESGSD I). Eur J Pediatr 161 Suppl 1:S20–34. doi: 10.1007/s00431-  
44  
45 002-0999-4  
46  
47

48  
49 Reddy SK, Austin SL, Spencer-Manzon M, et al. (2009) Liver transplantation for  
50  
51 glycogen storage disease type Ia. J Hepatol 51:483–490. doi: 10.1016/j.jhep.2009.05.026  
52

53  
54 Salganik SV, Weinstein DA, Shupe TD, et al. (2009) A detailed characterization of the  
55  
56 adult mouse model of glycogen storage disease Ia. Lab Invest J Tech Methods Pathol  
57  
58 89:1032–1042. doi: 10.1038/labinvest.2009.64  
59  
60  
61  
62  
63  
64  
65

1 Santer R, Hillebrand G, Steinmann B, Schaub J (2003) Intestinal glucose transport:  
2 evidence for a membrane traffic-based pathway in humans. *Gastroenterology* 124:34–39. doi:  
3  
4 10.1053/gast.2003.50009  
5  
6

7 Soty M, Chilloux J, Casteras S, et al. (2012) New insights into the organisation and  
8  
9 intracellular localisation of the two subunits of glucose-6-phosphatase. *Biochimie* 94:695–  
10  
11 703. doi: 10.1016/j.biochi.2011.09.022  
12  
13

14 Stümpel F, Burcelin R, Jungermann K, Thorens B (2001) Normal kinetics of intestinal  
15  
16 glucose absorption in the absence of GLUT2: evidence for a transport pathway requiring  
17  
18 glucose phosphorylation and transfer into the endoplasmic reticulum. *Proc Natl Acad Sci U S*  
19  
20 *A* 98:11330–11335. doi: 10.1073/pnas.211357698  
21  
22  
23

24 Visser G, Rake JP, Kokke FTM, et al. (2002) Intestinal function in glycogen storage  
25  
26 disease type I. *J Inherit Metab Dis* 25:261–267.  
27  
28

29 Wang DQ, Fiske LM, Carreras CT, Weinstein DA (2011) Natural history of  
30  
31 hepatocellular adenoma formation in glycogen storage disease type I. *J Pediatr* 159:442–446.  
32  
33 doi: 10.1016/j.jpeds.2011.02.031  
34  
35

36 Yiu WH, Lee YM, Peng W-T, et al. (2010) Complete normalization of hepatic G6PC  
37  
38 deficiency in murine glycogen storage disease type Ia using gene therapy. *Mol Ther J Am Soc*  
39  
40 *Gene Ther* 18:1076–1084. doi: 10.1038/mt.2010.64  
41  
42  
43

44 Yiu WH, Pan C-J, Ruef RA, et al. (2008) Angiotensin mediates renal fibrosis in the  
45  
46 nephropathy of glycogen storage disease type Ia. *Kidney Int* 73:716–723. doi:  
47  
48 10.1038/sj.ki.5002718  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
61  
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Table 1. The characteristics of mouse models of GSD1a.

	Total <i>G6pc</i> <sup>-/-</sup> mice	Liver <i>G6pc</i> <sup>-/-</sup> mice	Kidney <i>G6pc</i> <sup>-/-</sup> mice	Intestinal <i>G6pc</i> <sup>-/-</sup> mice
Glycaemia	≈40-50mg/dL in the fed state	Normal in the fed state (≈150-200 mg/dL) 50-60 mg/dL at 6h of fasting	Normal in the fed state Normal at 6h of fasting (≈140 mg/dL)	Normal
Plasmatic parameters	Hypertriglyceridemia (≈1.8-2.0 g/L) Hypercholesterolemia (≈1.8 g/L) Hyperuricemia (≈25 mg/L)  No or slight lactic acidemia	Hypertriglyceridemia (≈1.2 mg/L) Hypercholesterolemia (≈1.2 g/L) Hyperuricemia (≈25 mg/L)  Lactic acidemia	Normal TG  Normal cholesterol  Hyperuricemia (≈11 mg/L)  Normal lactic acid	Normal
Liver	Hepatomegaly (liver weight 10% BW) Steatosis	Hepatomegaly (liver weight: 8% of BW) Steatosis Late development of HCA*	Normal	Normal
Kidney	Nephromegaly (kidney weight: ≈3.5% BW)	Normal	Nephromegaly (kidney weight: ≈1.8% BW) Renal lipid accumulation Microalbuminuria Partial electrolyte imbalance	Normal
Intestine	Not determined	Normal	Normal	To be analyzed
Life expectancy	Need intensive glucose therapy or gene therapy to survive weaning	Normal	Over 12 months	Normal

The characteristics of 6-week-old total *G6pc*<sup>-/-</sup> mice were described by the Chou's team (Kim et al. 2008) (Lei et al. 1996). The data of liver *G6pc*<sup>-/-</sup> mice were obtained one month after *G6pc* deletion. \*The first hepatic lesions were observed 9 months after *G6pc* deletion (Mutel et al. 2011a). It is noteworthy that plasmatic parameters ameliorate with age in both GSD1a mouse models and patients. The data of kidney *G6pc*<sup>-/-</sup> mice were obtained 6 months after *G6pc* deletion (Clar et al. 2014). Intestinal *G6pc*<sup>-/-</sup> mice were analyzed up to 12 months after *G6pc* deletion (unpublished data). BW: body weight. Normal range of plasmatic values in 6 week-old C57Bl/6J mice: TG: 0.55-0.6 g/L; cholesterol: 0.7-0.8 g/L; Uric acid: 6-8 mg/L; liver and kidney weights represent about 4% and 1% of BW, respectively.

Table  
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Liver	Hepatomegaly (liver weight 10% BW) Steatosis	Hepatomegaly (liver weight: 8% of BW) Steatosis Late development of HCA*	Normal	Normal
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Intestine	Not determined	Normal	Normal	To be analyzed
Life expectancy	Need intensive glucose therapy or gene therapy to survive weaning	Normal	Over 12 months	Normal

Figure 1  
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