Gastroenterology 2016;150:574–580

REVIEWS IN BASIC AND CLINICAL GASTROENTEROLOGY AND HEPATOLOGY

Robert F. Schwabe and John W. Wiley, Section Editors

Circadian Clock Control of Liver Metabolic Functions

Hans Reinke[1,2] and Gad Asher[3]

[1]*Medical Faculty, Institute of Clinical Chemistry and Laboratory Diagnostics, University of Düsseldorf, Düsseldorf, Germany;* [2]*IUF-Leibniz Research Institute for Environmental Medicine, Düsseldorf, Germany; and* [3]*Department of Biological Chemistry, Weizmann Institute of Science, Rehovot, Israel*

The circadian clock is an endogenous biological timekeeping system that synchronizes physiology and behavior to day/night cycles. A wide variety of processes throughout the entire gastrointestinal tract and notably the liver appear to be under circadian control. These include various metabolic functions such as nutrient uptake, processing, and detoxification, which align organ function to cycle with nutrient supply and demand. Remarkably, genetic or environmental disruption of the circadian clock can cause metabolic diseases or exacerbate pathological states. In addition, modern lifestyles force more and more people worldwide into asynchrony between the external time and their circadian clock, resulting in a constant state of social jetlag. Recent evidence indicates that interactions between altered energy metabolism and disruptions in the circadian clock create a downward spiral that can lead to diabetes and other metabolic diseases. In this review, we provide an overview of rhythmic processes in the liver and highlight the functions of circadian clock genes under physiological and pathological conditions; we focus on their roles in regulation of hepatic glucose as well as lipid and bile acid metabolism and detoxification and their potential effects on the development of fatty liver and nonalcoholic steatohepatitis.

Keywords: Circadian Clock; Metabolism; Liver; Nonalcoholic Steatohepatitis.

Mammals have overt rest/activity and feeding/fasting cycles throughout the day. The resulting diurnal changes in nutrient supply and demand need to be handled primarily by the gastrointestinal tract and the liver, where nutrients are absorbed, processed, and directed to other organs in the body. The liver is a major metabolic hub; hepatic functions such as nutrient metabolism, detoxification, and synthesis of essential serum components must adapt to a rhythmically changing systemic environment. Like many other organs in the body, the liver has an internal timing system, the circadian clock, which adjusts physiological processes to their relevant time of day. The liver uses this system to anticipate recurring systemic and environmental changes and function in a proactive manner. Numerous studies have used the liver as a prototype for identifying general principles of core clock circuitry. We review the evidence to support the circadian control of liver metabolic functions.

Virtually all metabolic activities in the gastrointestinal tract and the liver have daily rhythms regulated by the clock, food, or both. In fact, food- and clock-regulated processes interact and in many cases cannot be readily uncoupled. Several recent reviews have addressed circadian control mechanisms of different parts of the gastrointestinal tract (the stomach and small and large intestine[1–4]). In this review, we focus on the various metabolic functions of the liver that are under circadian control and thereby are likely to support proper liver and whole body homeostasis.

Molecular and Anatomic Organization

The circadian clock is a cell-autonomous molecular mechanism that is organized in a hierarchical structure on the organismal level. Cellular circadian oscillators confer rhythmic expression to large numbers of genes, which leads to overt daily changes in physiology and behavior.[5] The core of the molecular mammalian clock comprises a negative feedback loop of period (*Per*) and cryptochrome (*Cry*) gene expression (Figure 1). Expression of *Per* and *Cry* genes is activated by the transcription factors ARNTL (also known as BMAL1) and CLOCK. Over time, period and cryptochrome proteins accumulate in the cell and ultimately repress transcription at their own gene loci. The core oscillator is stabilized by nuclear receptors of the ROR (RORA, RORB, RORG) and NR1D1 (also known as REV-ERBA) and NR1D2 (also known as REV-ERBB) types, which regulate expression of *Bmal1*. Clocks are fine-tuned by posttranscriptional regulation of these gene products.[6]

Mammalian oscillators are structured hierarchically on the anatomic level. A master clock in the brain sets the time for all other body clocks. This central pacemaker resides in the suprachiasmatic nucleus (SCN) in the hypothalamus and is synchronized with the geophysical time by the zeitgeber (time giver) light via the retinohypothalamic tract. The SCN uses neuronal and humoral pathways to transmit temporal information to peripheral organs in the rest of the body.[7] Feeding cycles appear to serve as the dominant zeitgeber for clocks in peripheral organs,[8] which supports the idea that these clocks are highly responsive to metabolic cues.[9,10]

Abbreviation used in this paper: SCN, suprachiasmatic nucleus.

Most current article

0016-5085/$36.00
http://dx.doi.org/10.1053/j.gastro.2015.11.043

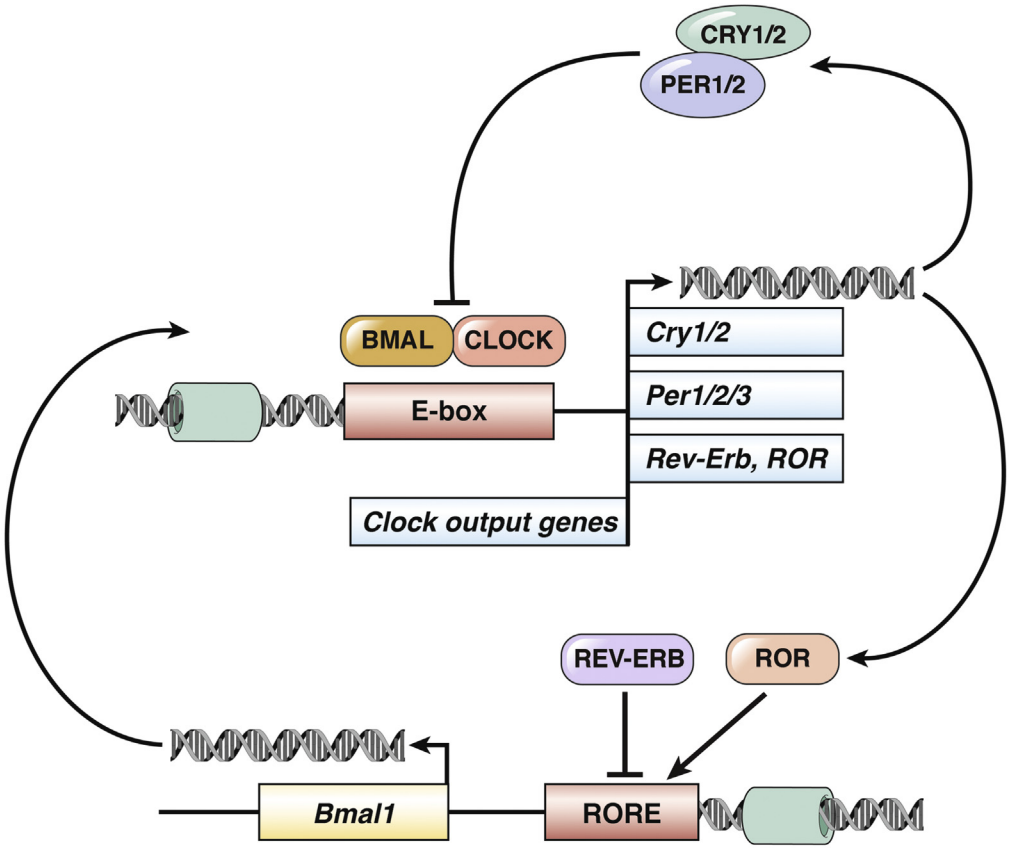

Figure 1. The core circadian oscillator. The activator proteins CLOCK and BMAL1 control the expression of *Per*, *Cry*, *Nr1d*, and *Ror* genes via E-box elements in their promoter regions. PER and CRY proteins repress their own transcription, generating a transcriptional and translational feedback loop. NR1D and ROR proteins form an auxiliary feedback loop by regulating *Bmal1* expression.

Accordingly, the molecular clockwork has been linked to the cellular redox metabolism via a reduced nicotinamide adenine dinucleotide (NADH)–nicotinamide phosphoribosyltransferase (NAMPT) feedback loop[11,12] with connections to SIRT proteins,[13–17] PARP1,[18] and evolutionarily conserved redox oscillators.[19,20] In particular, peripheral oscillators such as the liver clock are additionally coupled to cellular and organismal physiology through energy and nutrient sensing systems such as AMP-activated protein kinase (AMPK),[21] PPARGC1A (also known as PGC1),[22] metabolic feedback loops involving metabolites such as polyamines,[23] and nuclear hormone receptor signaling pathways.[24] They also sense physical parameters such as body temperature and use this information to fine-tune their functions in an organ-specific manner.[25–27] Thus, in contrast to the master clock, for which light is the dominant zeitgeber, peripheral clocks are strongly affected by cell metabolism, the physiological and metabolic state of the surrounding tissue, and serum-borne signals[28]; they can be efficiently entrained by feeding/fasting rhythms to the point of being fully uncoupled from SCN rhythms.[8]

Circadian Liver Functions

Oscillations of the core circadian clock have to be transmitted to transcriptional signals to control rhythmic output. For this purpose, many rate-limiting metabolic enzymes are under the direct control of core clock transcription factors. More frequently, however, the core clock controls the rhythmic expression of auxiliary, often organ-specific, transcription factors that in turn control the cyclic expression of different enzymes and metabolic master regulators.

Genome-wide gene expression studies provided the first and important insights into the role of the circadian clock in global liver physiology.[29–32] In mouse liver, 2 peaks of rhythmically regulated transcripts are observed at the end of the light and dark phases, respectively, likely reflecting the highly differential physiological requirements, such as in energy demand or detoxification activity, mediated by activity or rest. The circadian expression phases of many rhythmic messenger RNAs are in accordance with the phases of the proteins they encode and their respective biochemical pathways. It should be noted, however, that recent proteomic studies[33,34] found that some metabolic enzymes cycle whereas their transcripts are relatively constant throughout the day, indicating that posttranscriptional mechanisms are also involved in circadian regulation of liver functions. Recent analyses of the liver metabolome further support this idea.[35,36]

The first set of global transcriptome analyses[29–31] revealed that most principal functions of the liver are rhythmically regulated, as exemplified for the following metabolic pathways (Figure 2). The liver controls energy homeostasis, and many rate-limiting enzymes in nutrient metabolism are

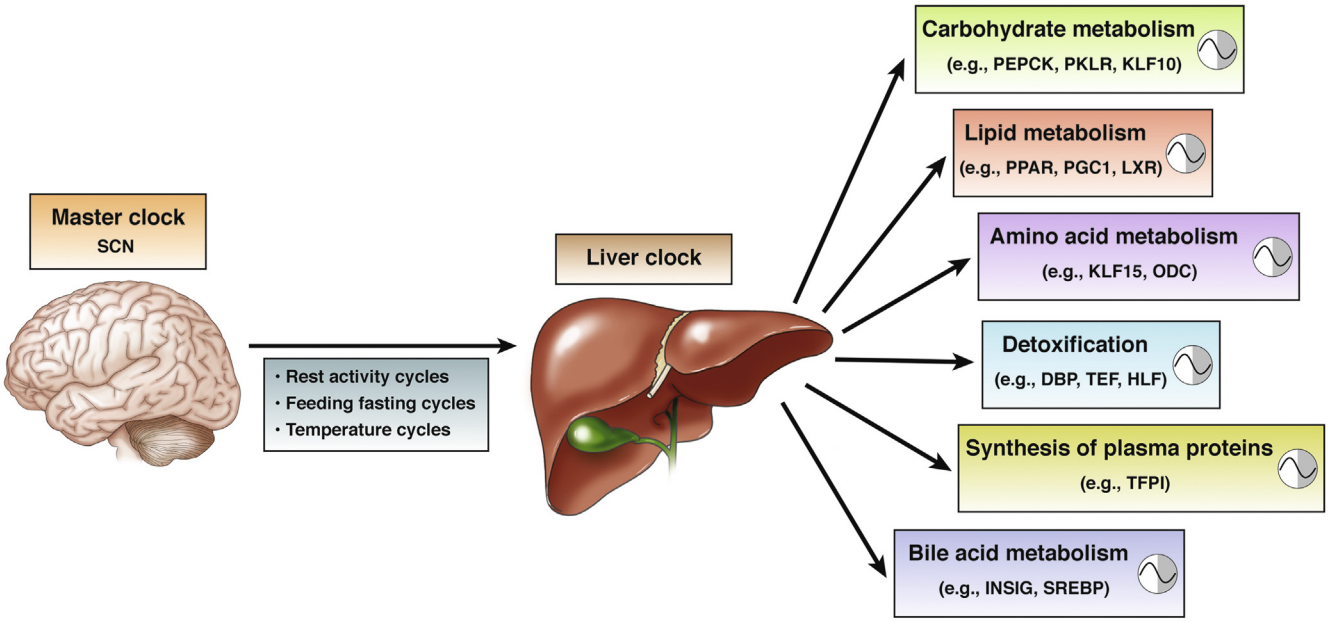

Figure 2. Circadian regulation of liver physiology. The hypothalamic master clock synchronizes peripheral oscillators, such as the liver clock, via rhythmic activity, feeding, and temperature cues. The liver clock drives the cyclic expression of master regulators and rate-limiting enzymes of key hepatic metabolic outputs.

expressed in a circadian manner. For example, the expression levels of glucose transporters, the glucagon receptor, and other enzymes regulating the metabolism of hexose sugars were observed with peak phases of expression in the early evening, when animals ingest most of their daily food ration. In regard to lipid homeostasis, multiple enzymes within the glycerol 3-phosphate pathway (such as glycerol-3-phosphate acyltransferase [GPAT], 1-acyl-glycerol-3-phosphate acyltransferase [AGPAT], and LPIN), which regulates glycerol and lipid metabolism and triglyceride accumulation, are expressed in a circadian manner.[36]

The liver also metabolizes cholesterol and steroid hormones. Expression of 3-hydroxy-3-methylglutaryl–coenzyme A (HMG-CoA) reductase, a cholesterol biosynthetic protein, peaks at a time of day when cholesterol is not supplied from the diet, whereas the expression phases of P450 and various cytochromes involved in the breakdown of cholesterol and steroid hormones are more widely distributed. The liver is the main site for the production of active thyroxine hormone, which is controlled by cycling hepatic expression of deiodinase 1, which rhythmically deiodinizes inactive T4 thyroxine to the active T3 form. Furthermore, the expression phase of the gene for the thyroid hormone receptor α is aligned with the circadian rhythm of T3 and T4 serum levels. Also, transcript levels of innate immunity proteins, many of which are synthesized in liver, have circadian rhythms. As a last example, the liver is a major site for the synthesis of coagulation and fibrinolytic proteins, and tissue factor pathway inhibitor 2, which has a central role in the inhibition of TF-VIIa complex formation, is rhythmically induced in the liver. More globally, uptake and secretion of small molecules in the liver is in many cases regulated in a circadian manner via the rhythmic expression of membrane channels and transporters.[29–31]

Subsequent studies revealed that rhythmic hepatic gene expression seems to be controlled through many interacting cellular and systemic circadian mechanisms. Expression of genes that control the circadian clock can by regulated directly by the cellular clock, by rhythmic systemic signals that originate, for example, from periodic food ingestion, or by a combination of both mechanisms.[37,38] It is conceivable that the circadian component of this regulation optimizes the anticipation of predicable fluctuations in the organ environment, whereas the responsiveness to systemic cues adds a degree of flexibility to the system that is necessary to adapt to unforeseeable changes of environmental conditions (eg, in times of food scarcity).

Regulation of Glucose Homeostasis

The liver has an important role in regulation of glucose homeostasis, along with the pancreas, brain, and skeletal muscle. Although direct glucose signaling is the main process by which the body adapts to rapid changes in glucose availability, the circadian clock seems to provide rhythmic baseline regulation to regularly recurring events, such as food uptake after nocturnal starvation.[39] Accordingly, many related processes, such as insulin and glucagon secretion[40,41] as well as glucose production and uptake,[42,43] have a circadian component that is clearly discernible from nutrient signaling. Rhythmic insulin secretion is controlled by pancreatic clocks in rodents[44] and humans,[45] which seem to be indispensable for pancreatic function. The circadian clocks in the SCN and the liver use different mechanisms to generate antiphasic rhythms of glucose metabolism, which in combination produce nearly constant blood levels of glucose throughout the day.[39] The master pacemaker in the brain controls rest/activity and feeding/fasting rhythms, which lead to rhythmic

nutrient uptake and signaling. The main role of the liver clock is to buffer the circadian fluctuations of blood glucose levels that originate from these behavioral cycles.

The importance of this mechanism was revealed by targeted disruption of the liver clock. Disruption of the essential clock gene *Bmal1* in livers of mice led to excessive fluctuations in blood glucose levels, mainly in the post-absorptive phase, controlled by the now-dominant clock in the SCN.[39] In contrast, loss of clock function in the whole body resulted in only slight defects in glucose tolerance and gluconeogenesis after insulin-induced hypoglycemia but otherwise normal glucose levels throughout the day.[39,46]

Several core clock and clock-controlled genes have additional roles in glucose metabolism. Cryptochromes regulate hepatic gluconeogenesis through interaction with G protein–coupled receptors, which blocks accumulation of adenosine 3′,5′-cyclic monophosphate (cAMP) and activation of transcription of gluconeogenic genes regulated by CREB.[47] Importantly, the same study showed that overexpression of *Cry1* specifically in the liver lowers blood glucose levels and increases insulin sensitivity in diabetic mice. In an alternative pathway, cryptochromes repress transcription of the genes encoding the glucocorticoid receptor and phosphoenolpyruvate carboxykinase, which regulates gluconeogenesis.[48] Glucocorticoids also affect glucose metabolism by inducing expression of *Per2* under hyperglycemic conditions; mice lacking an essential glucocorticoid response element in the *Per2* gene are protected from glucose intolerance provoked by prolonged glucocorticoid treatment.[49]

Interestingly, KLF10, a transcription factor regulated by CLOCK and BMAL1, links the circadian clock and metabolism in a sex-specific manner. In mouse liver, KLF10 regulates the expression of genes involved in glycolysis and gluconeogenesis. Loss of KLF10 from male mice led to postprandial and fasting hyperglycemia, whereas female mice remained normoglycemic.[50] Notably, another member of the Klf transcription factor family, KLF15, is believed to regulate rhythmic expression of multiple enzymes involved in hepatic nitrogen and amino acid homeostasis.[51]

In summary, clock-dependent regulation of glucose metabolism is controlled by positive and negative regulators of the core clock oscillator. From there, multiple signaling pathways converge on rate-limiting enzymes of glucose anabolism and catabolism, such as phosphoenolpyruvate carboxykinase or pyruvate kinase.[47,50]

Control of Lipid and Bile Acid Metabolism

The liver participates in lipid metabolism by regulating lipoprotein synthesis and lipid uptake and conversion, as well as de novo synthesis and oxidation of fatty acids. The circadian clock regulates most aspects of hepatic lipid metabolism; conversely, lipids are potential regulators of circadian rhythmicity.[52] A recent lipidomic analysis of mice that do (control) vs do not express CLOCK provided extensive insight into the rhythmic accumulation of lipids in the liver. Surprisingly, a similar fraction of all lipids (approximately 17%) oscillated diurnally in mice with and without CLOCK. However, the composition and circadian phase differed in CLOCK-null mice. Moreover, when control mice were fed only during the night, the circadian phase of triglyceride accumulation shifted and hepatic triglyceride levels decreased by 50%, without changes in total caloric intake. Consequently, the combination of circadian clock–dependent regulation and feeding time determines circadian oscillations of hepatic triglycerides, which can nevertheless persist in the absence of a functional clock.[36]

Circadian lipid metabolism is not surprisingly controlled to a large extent by the clock-dependent regulation of key enzymes and transcription factors. Among others, the biosynthetic enzymes in the glycerol 3-phosphate pathway, an important pathway of triglyceride biosynthesis in the liver, together with enzymes that regulate fatty acid synthesis, such as ELOVL3, ELOVL6, and FAS, have rhythmic expression patterns. Likewise, lipid regulatory factors such as peroxisome proliferator-activated receptor (PPAR), PGC1, SREBP1, NR1D2, and ROR are expressed in rhythmic patterns in liver cells.[52,53] The expression of several lipid metabolism genes is altered in mice with disruption of *Clock*, which results in dysregulated accumulation of intermediates and products related to lipid metabolism in the liver.[35] Similarly, mice lacking the *Per2* gene have dyslipidemia.[54]

Another core clock protein, NR1D1, represses expression of *Apoc3* messenger RNA in the liver. Consequently, serum levels of APOC3 and very-low-density lipoprotein triglycerides, which are risk factors for atherosclerosis, are increased in mice with disruption of *Nr1d1*.[55] Disruption of the circadian clock can also lead to hepatic steatosis, which has been shown in mice with disruptions in *Clock*[56] and *Nr1d1-Nr1d2*[57] or on deletion of the histone deacetylase HDAC3, which is recruited by NR1D1 to genes that regulate lipid metabolism.[58] In contrast, restricted feeding can protect mice from hepatic steatosis.[59]

Indirect evidence for the control of circadian clock functions by lipids mainly stems from studies of nuclear receptors that are embedded within the core oscillator mechanism. RORA, RORG, and PPAR (A, G, and D), which are all expressed in the liver, are an integral part of the core clock by regulating *Bmal1* transcription and at the same time participating in the control of lipid metabolism. Importantly, cholesterol and certain oxysterols regulate the transcriptional activation potential of RORA and RORG, whereas PPAR isoforms are bound by various fatty acids and eicosanoids.[52] Moreover, SIRT6, a chromatin modifier involved in hepatic clock gene transcription as well as SREBP-dependent regulation of fatty acid and cholesterol metabolism,[17] is activated by long-chain fatty acids.[60] Taken together, these studies provide evidence for a metabolic feedback loop within the core clock mechanism involving lipid metabolic intermediates.

The liver is the principal organ for the conversion of cholesterol into bile acids, which facilitate the absorption of nutrients in the intestine and have paracrine and endocrine functions. Bile acid homeostasis is principally governed by a

feedback loop involving FXR, FGF15, and SHP,[61] but similarly to the homeostatic regulation of systemic glucose levels, the circadian clock provides additional regulatory layers. NR1D1 promotes circadian signaling via INSIG2–SREBP and LXR, which promotes rhythmic expression of the rate-limiting enzyme cholesterol 7a-hydroxylase CYP7A1 and other genes involved in cholesterol and lipid metabolism.[53] Additionally, the PAR-domain basic leucine zipper (PAR bZIP) protein DBP rhythmically activates expression of CYP7A1 in the liver.[62] The cycling transcription factor KLF15, which functions as a repressor of the FXR–FGF15 signaling pathway, also controls bile acid synthesis.[63] Notably, diurnal rhythms of bile acid synthesis have also been shown in humans.[64]

Rhythmic Detoxification in the Liver

The liver also functions to clear toxic substances from the blood. Hepatic detoxification is separated into different phases, which are under individual control of the circadian clock. In general, toxins or xenobiotics are first transformed into water-soluble metabolites and then excreted from the body.

The first step in detoxification often involves the binding of xenobiotics to nuclear receptors, followed by transcriptional activation of detoxification pathways. Circadian regulation in this step is controlled by rhythmic expression levels of nuclear receptor genes in the liver and other tissues.[65] Phase 1 detoxification involves substrate oxidation by various cytochromes, which are, together with other phase 1 proteins, regulated in a circadian manner in the liver; expression peaks at the time when animals ingest food and have the highest chance for exposure to foodborne toxins.[66]

In phase 2 of the detoxification process, toxins are rendered hydrophilic by conjugating enzymes to make them excretable. These enzymes are also rhythmically expressed, but in contrast to the phase 1 enzymes, their expression peaks in widely different circadian phases.[66] Excretion is initiated by transporter proteins of various classes in phase 3 detoxification.

In the liver, toxins are excreted into the bile by membrane transporters, although this step has weak circadian regulation. Master regulators of all classes of detoxification enzymes are the liver-specific PAR bZIP proteins DBP, TEF, and HLF, which are rhythmically activated through CLOCK and BMAL1 binding sites in their promoters. Mice that lack all 3 PAR bZIP proteins have premature aging syndromes and deficits in basal and inducible hepatic detoxification due to widespread dysregulation of phase I and II detoxification enzymes and regulators, such as the nuclear receptor CAR.[67]

Effects on Fatty Liver, Nonalcoholic Steatohepatitis, and the Microbiome

Clock-related research might also lead to ways to prevent or treat nonalcoholic fatty liver disease, a major public health challenge. The circadian clock is involved in regulation of hepatic triglyceride accumulation, inflammation, oxidative stress, and mitochondrial dysfunction,[36,59,68] which contribute to the pathogenesis of nonalcoholic steatohepatitis—the advanced form of nonalcoholic fatty liver disease.[69] Intervention strategies targeted at circadian oscillator components[70] might therefore be used for treatment of liver diseases. Even more importantly, circadian misalignment has been identified as a risk factor for metabolic disease.[71] Behavioral modification is widely seen as a powerful tool in the battle against socioeconomic diseases, such as the various manifestations of nonalcoholic fatty liver disease and their comorbidities, so strategies to optimize rotating shift work schedules and to reduce social jetlag might improve the health of the entire population.

A recent series of studies analyzed the rhythmicity of the intestinal microbiome and found that it affected metabolic homeostasis in the gastrointestinal tract.[72–75] More specifically, feeding time was shown to shape the rhythmicity and composition of the gut microbiota, raising the possibility that daily rhythms in bile acid synthesis also take part in this process. Disruptions of the intestinal microbiome have been associated with diet, obesity, and metabolic disease and might be reversed, so circadian control of the microbiota will be an interesting area of research with potential relevance to fatty liver and nonalcoholic steatohepatitis.

References

1. Hussain MM. Regulation of intestinal lipid absorption by clock genes. Annu Rev Nutr 2014;34:357–375.
2. Hoogerwerf WA. Role of biological rhythms in gastrointestinal health and disease. Rev Endocr Metab Disord 2009;10:293–300.
3. Konturek PC, Brzozowski T, Konturek SJ. Gut clock: implication of circadian rhythms in the gastrointestinal tract. J Physiol Pharmacol 2011;62:139–150.
4. Asher G, Sassone-Corsi P. Time for food: the intimate interplay between nutrition, metabolism, and the circadian clock. Cell 2015;161:84–92.
5. Dibner C, Schibler U. Circadian timing of metabolism in animal models and humans. J Intern Med 2015; 277:513–527.
6. Brown SA, Kowalska E, Dallmann R. (Re)inventing the circadian feedback loop. Dev Cell 2012;22:477–487.
7. Welsh DK, Takahashi JS, Kay SA. Suprachiasmatic nucleus: cell autonomy and network properties. Annu Rev Physiol 2010;72:551–577.
8. **Damiola F, Le Minh N**, Preitner N, et al. Restricted feeding uncouples circadian oscillators in peripheral tissues from the central pacemaker in the suprachiasmatic nucleus. Genes Dev 2000;14:2950–2961.
9. Green CB, Takahashi JS, Bass J. The meter of metabolism. Cell 2008;134:728–742.
10. Asher G, Schibler U. Crosstalk between components of circadian and metabolic cycles in mammals. Cell Metab 2011;13:125–137.
11. **Ramsey KM, Yoshino J, Brace CS**, et al. Circadian clock feedback cycle through NAMPT-mediated NAD+ biosynthesis. Science 2009;324:651–654.
12. Nakahata Y, Sahar S, Astarita G, et al. Circadian control of the NAD+ salvage pathway by CLOCK-SIRT1. Science 2009;324:654–657.

13. Asher G, Gatfield D, Stratmann M, et al. SIRT1 regulates circadian clock gene expression through PER2 deacetylation. Cell 2008;134:317–328.
14. **Nakahata Y, Kaluzova M**, Grimaldi B, et al. The NAD+-dependent deacetylase SIRT1 modulates CLOCK-mediated chromatin remodeling and circadian control. Cell 2008;134:329–340.
15. Chang HC, Guarente L. SIRT1 mediates central circadian control in the SCN by a mechanism that decays with aging. Cell 2013;153:1448–1460.
16. **Peek CB, Affinati AH**, Ramsey KM, et al. Circadian clock NAD+ cycle drives mitochondrial oxidative metabolism in mice. Science 2013;342:1243417.
17. Masri S, Rigor P, Cervantes M, et al. Partitioning circadian transcription by SIRT6 leads to segregated control of cellular metabolism. Cell 2014;158:659–672.
18. Asher G, Reinke H, Altmeyer M, et al. Poly(ADP-Ribose) Polymerase 1 participates in the phase entrainment of circadian clocks to feeding. Cell 2010;142:943–953.
19. O'Neill JS, Reddy AB. Circadian clocks in human red blood cells. Nature 2011;469:498–503.
20. **O'Neill JS, van Ooijen G**, Dixon LE, et al. Circadian rhythms persist without transcription in a eukaryote. Nature 2011;469:554–558.
21. **Lamia KA, Sachdeva UM**, DiTacchio L, et al. AMPK regulates the circadian clock by cryptochrome phosphorylation and degradation. Science 2009;326:437–440.
22. Liu C, Li S, Liu T, et al. Transcriptional coactivator PGC-1alpha integrates the mammalian clock and energy metabolism. Nature 2007;447:477–481.
23. Zwighaft Z, Aviram R, Shalev M, et al. Circadian clock control by polyamine levels through a mechanism that declines with age. Cell Metab 2015;22:874–885.
24. Teboul M, Guillaumond F, Grechez-Cassiau A, et al. The nuclear hormone receptor family round the clock. Mol Endocrinol 2008;22:2573–2582.
25. Buhr ED, Yoo SH, Takahashi JS. Temperature as a universal resetting cue for mammalian circadian oscillators. Science 2010;330:379–385.
26. Reinke H, Saini C, Fleury-Olela F, et al. Differential display of DNA-binding proteins reveals heat-shock factor 1 as a circadian transcription factor. Genes Dev 2008;22:331–345.
27. Saini C, Morf J, Stratmann M, et al. Simulated body temperature rhythms reveal the phase-shifting behavior and plasticity of mammalian circadian oscillators. Genes Dev 2012;26:567–580.
28. Gerber A, Esnault C, Aubert G, et al. Blood-borne circadian signal stimulates daily oscillations in actin dynamics and SRF activity. Cell 2013;152:492–503.
29. **Panda S, Antoch MP**, Miller BH, et al. Coordinated transcription of key pathways in the mouse by the circadian clock. Cell 2002;109:307–320.
30. Storch KF, Lipan O, Leykin I, et al. Extensive and divergent circadian gene expression in liver and heart. Nature 2002;417:78–83.
31. **Akhtar RA, Reddy AB**, Maywood ES, et al. Circadian cycling of the mouse liver transcriptome, as revealed by cDNA microarray, is driven by the suprachiasmatic nucleus. Curr Biol 2002;12:540–550.
32. **Zhang R, Lahens NF**, Ballance HI, et al. A circadian gene expression atlas in mammals: implications for biology and medicine. Proc Natl Acad Sci U S A 2014;111:16219–16224.
33. Robles MS, Cox J, Mann M. In-vivo quantitative proteomics reveals a key contribution of post-transcriptional mechanisms to the circadian regulation of liver metabolism. PLoS Genet 2014;10:e1004047.
34. Mauvoisin D, Wang J, Jouffe C, et al. Circadian clock-dependent and -independent rhythmic proteomes implement distinct diurnal functions in mouse liver. Proc Natl Acad Sci U S A 2014;111:167–172.
35. Eckel-Mahan KL, Patel VR, Mohney RP, et al. Coordination of the transcriptome and metabolome by the circadian clock. Proc Natl Acad Sci U S A 2012;109:5541–5546.
36. Adamovich Y, Rousso-Noori L, Zwighaft Z, et al. Circadian clocks and feeding time regulate the oscillations and levels of hepatic triglycerides. Cell Metab 2014;19:319–330.
37. Kornmann B, Schaad O, Bujard H, et al. System-driven and oscillator-dependent circadian transcription in mice with a conditionally active liver clock. PloS Biol 2007;5:e34.
38. Vollmers C, Gill S, DiTacchio L, et al. Time of feeding and the intrinsic circadian clock drive rhythms in hepatic gene expression. Proc Natl Acad Sci U S A 2009;106:21453–21458.
39. Lamia KA, Storch KF, Weitz CJ. Physiological significance of a peripheral tissue circadian clock. Proc Natl Acad Sci U S A 2008;105:15172–15177.
40. Ruiter M, La Fleur SE, van Heijningen C, et al. The daily rhythm in plasma glucagon concentrations in the rat is modulated by the biological clock and by feeding behavior. Diabetes 2003;52:1709–1715.
41. Boden G, Ruiz J, Urbain JL, et al. Evidence for a circadian rhythm of insulin secretion. Am J Physiol 1996;271:E246–E252.
42. la Fleur SE, Kalsbeek A, Wortel J, et al. A daily rhythm in glucose tolerance: a role for the suprachiasmatic nucleus. Diabetes 2001;50:1237–1243.
43. Bolli GB, De Feo P, De Cosmo S, et al. Demonstration of a dawn phenomenon in normal human volunteers. Diabetes 1984;33:1150–1153.
44. Marcheva B, Ramsey KM, Buhr ED, et al. Disruption of the clock components CLOCK and BMAL1 leads to hypoinsulinaemia and diabetes. Nature 2010;466:627–631.
45. Pulimeno P, Mannic T, Sage D, et al. Autonomous and self-sustained circadian oscillators displayed in human islet cells. Diabetologia 2013;56:497–507.
46. **Rudic RD, McNamara P**, Curtis AM, et al. BMAL1 and CLOCK, two essential components of the circadian clock, are involved in glucose homeostasis. PLoS Biol 2004;2:e377.
47. **Zhang EE, Liu Y**, Dentin R, et al. Cryptochrome mediates circadian regulation of cAMP signaling and hepatic gluconeogenesis. Nat Med 2010;16:1152–1156.

48. Lamia KA, Papp SJ, Yu RT, et al. Cryptochromes mediate rhythmic repression of the glucocorticoid receptor. Nature 2011;480:552–556.
49. So AY, Bernal TU, Pillsbury ML, et al. Glucocorticoid regulation of the circadian clock modulates glucose homeostasis. Proc Natl Acad Sci U S A 2009;106:17582–17587.
50. Guillaumond F, Grechez-Cassiau A, Subramaniam M, et al. Kruppel-like factor KLF10 is a link between the circadian clock and metabolism in liver. Mol Cell Biol 2010;30:3059–3070.
51. Jeyaraj D, Scheer FA, Ripperger JA, et al. Klf15 orchestrates circadian nitrogen homeostasis. Cell Metab 2012;15:311–323.
52. Adamovich Y, Aviram R, Asher G. The emerging roles of lipids in circadian control. Biochim Biophys Acta 2015;1851:1017–1025.
53. Le Martelot G, Claudel T, Gatfield D, et al. REV-ERBalpha participates in circadian SREBP signaling and bile acid homeostasis. PloS Biol 2009;7:e1000181.
54. Grimaldi B, Bellet MM, Katada S, et al. PER2 controls lipid metabolism by direct regulation of PPARgamma. Cell Metab 2010;12:509–520.
55. **Raspe E, Duez H**, Mansen A, et al. Identification of Rev-erbalpha as a physiological repressor of apoC-III gene transcription. J Lipid Res 2002;43:2172–2179.
56. Turek FW, Joshu C, Kohsaka A, et al. Obesity and metabolic syndrome in circadian Clock mutant mice. Science 2005;308:1043–1045.
57. Bugge A, Feng D, Everett LJ, et al. Rev-erbalpha and Rev-erbbeta coordinately protect the circadian clock and normal metabolic function. Genes Dev 2012;26:657–667.
58. **Feng D, Liu T**, Sun Z, et al. A circadian rhythm orchestrated by histone deacetylase 3 controls hepatic lipid metabolism. Science 2011;331:1315–1319.
59. **Hatori M, Vollmers C, Zarrinpar A**, et al. Time-restricted feeding without reducing caloric intake prevents metabolic diseases in mice fed a high-fat diet. Cell Metab 2012;15:848–860.
60. Feldman JL, Baeza J, Denu JM. Activation of the protein deacetylase SIRT6 by long-chain fatty acids and widespread deacylation by mammalian sirtuins. J Biol Chem 2013;288:31350–31356.
61. Lu TT, Makishima M, Repa JJ, et al. Molecular basis for feedback regulation of bile acid synthesis by nuclear receptors. Mol Cell 2000;6:507–515.
62. Lavery DJ, Schibler U. Circadian transcription of the cholesterol 7 alpha hydroxylase gene may involve the liver-enriched bZIP protein DBP. Genes Dev 1993;7:1871–1884.
63. **Han SS, Zhang R**, Jain R, et al. Circadian control of bile acid synthesis by a KLF15-Fgf15 axis. Nat Commun 2015;6:7231.
64. Duane WC, Levitt DG, Mueller SM, et al. Regulation of bile acid synthesis in man. Presence of a diurnal rhythm. J Clin Invest 1983;72:1930–1936.
65. Yang X, Downes M, Yu RT, et al. Nuclear receptor expression links the circadian clock to metabolism. Cell 2006;126:801–810.
66. Zhang YK, Yeager RL, Klaassen CD. Circadian expression profiles of drug-processing genes and transcription factors in mouse liver. Drug Metab Dispos 2009;37:106–115.
67. Gachon F, Olela FF, Schaad O, et al. The circadian PAR-domain basic leucine zipper transcription factors DBP, TEF, and HLF modulate basal and inducible xenobiotic detoxification. Cell Metab 2006;4:25–36.
68. **Jacobi D, Liu S**, Burkewitz K, et al. Hepatic Bmal1 regulates rhythmic mitochondrial dynamics and promotes metabolic fitness. Cell Metab 2015;22:709–720.
69. Dowman JK, Tomlinson JW, Newsome PN. Pathogenesis of non-alcoholic fatty liver disease. QJM 2010;103:71–83.
70. **Solt LA, Wang Y**, Banerjee S, et al. Regulation of circadian behaviour and metabolism by synthetic REV-ERB agonists. Nature 2012;485:62–68.
71. Parsons MJ, Moffitt TE, Gregory AM, et al. Social jetlag, obesity and metabolic disorder: investigation in a cohort study. Int J Obes (Lond) 2015;39:842–848.
72. Liang X, Bushman FD, FitzGerald GA. Rhythmicity of the intestinal microbiota is regulated by gender and the host circadian clock. Proc Natl Acad Sci U S A 2015;112:10479–10484.
73. Leone V, Gibbons SM, Martinez K, et al. Effects of diurnal variation of gut microbes and high-fat feeding on host circadian clock function and metabolism. Cell Host Microbe 2015;17:681–689.
74. Thaiss CA, Zeevi D, Levy M, et al. Transkingdom control of microbiota diurnal oscillations promotes metabolic homeostasis. Cell 2014;159:514–529.
75. Mukherji A, Kobiita A, Ye T, et al. Homeostasis in intestinal epithelium is orchestrated by the circadian clock and microbiota cues transduced by TLRs. Cell 2013;153:812–827.

Author names in bold designate shared co-first authorship.

Received August 24, 2015. Accepted November 3, 2015.

Reprint requests
Address requests for reprints to: Dr Hans Reinke, Medical Faculty, Institute of Clinical Chemistry and Laboratory Diagnostics, University of Düsseldorf, 40225 Düsseldorf, Germany. e-mail: hans.reinke@med.uni-duesseldorf.de; and Gad Asher, MD, PhD, Department of Biological Chemistry, Weizmann Institute of Science, Rehovot, 7610001, Israel. e-mail: gad.asher@weizmann.ac.il.

Acknowledgments
The authors apologize to all colleagues whose work could not be cited due to space limitations.

Conflicts of interest
The authors disclose no conflicts.

Funding
Supported by the German-Israeli Foundation for Scientific Research and Development (G-1199-230.9/2012), the Deutsche Forschungsgemeinschaft (RE 3046/2-1 to H.R.), the Israel Science Foundation (ISF 138/12 to G.A.), the HFSP Career Development Award (HFSP CDA00014/2012 to G.A.), and the European Research Council (ERC-2011 METACYCLES 310320 to G.A.).