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14. ABSTRACT

Data generated during this year has uncovered a role for SIRT6 as a critical regulator of ISC activity. Using two different mouse models as well as an *in vitro* intestinal organoid system, I have found that lack of SIRT6 increases the number and activity of ISCs, a phenotype that is reversed by inhibiting glycolysis, indicating that enhanced glycolytic metabolism in the absence of SIRT6 drives intestinal tumorigenesis by increasing the number of tumor-initiating cells. In addition, we have found that ISCs are more glycolytic than more differentiated intestinal epithelial cells, and increased glycolysis in these cells regulates their stemness. Taken together, these results are in agreement with a model in which SIRT6 regulates ISCs activity by controlling glucose metabolism and, when lost, increased glycolysis promotes ISC expansion and the generation of potential tumor-initiating cells.

15. SUBJECT TERMS
SIRT6, intestinal tumorigenesis, cancer metabolism, intestinal stem cells, tumor initiating cells

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1. INTRODUCTION

Colorectal carcinoma (CRC) is the third leading cause of cancer morbidity and mortality in the United States. Familial Adenomatous Polyposis (FAP) represents one of the most common syndromes associated with high penetrant hereditary CRC. A prominent feature of cancer cells is their increased glucose uptake and reliance on aerobic glycolytic metabolism, a phenomenon described by Otto Warburg decades ago. Though it is a potential candidate for targeting against tumors, little is known about the mechanisms controlling it. Remarkably, we have recently identified the SIRT6 histone deacetylase as a central regulator of glycolytic metabolism: cells lacking SIRT6 undergo a dramatic metabolic switch, increasing glycolysis and inhibiting mitochondrial respiration (Mostoslavsky et al., 2006; Zhong et al., 2010). In this proposal, we aimed to study the role of SIRT6 in colorectal cancer cells. We hypothesize that colon cancer cells might selectively down-modulate SIRT6 to acquire a selective advantage in order to grow under conditions of glycolytic metabolism. Indeed, our work done over these two years indicate that loss of SIRT6 provides tumorigenic potential to otherwise normal cells, modulating glycolysis and bypassing classical oncogenic pathways (Sebastian et al., 2012). Furthermore, we have established SIRT6 as a critical tumor suppressor in intestinal tumorigenesis by regulating cancer metabolism (Sebastian et al., 2012). Data generated during this year has expanded these observations and uncovered a new role for SIRT6 and glucose metabolism as regulators of intestinal stem cell activity and tumor initiation in intestinal cancer. Overall, our results provide new insights into the molecular mechanisms regulating colon cancer metabolism. In this context, modulation of SIRT6 activity could provide us in the future with a potential therapeutic approach to tackle cancer development.

2. KEYWORDS

SIRT6, cancer metabolism, aerobic glycolysis, intestinal tumorigenesis, intestinal stem cells, tumor initiating cells.

3. OVERALL PROJECT SUMMARY

Our work done during the first year demonstrated that SIRT6 suppresses intestinal tumorigenesis *in vivo* by inhibiting glucose metabolism reprogramming (Sebastian et al., 2012). Importantly, lack of SIRT6 increases the number of tumors in the intestine of APCmin mice, suggesting a role for this chromatin factor in tumor initiation. Work done by Hans Clevers laboratory

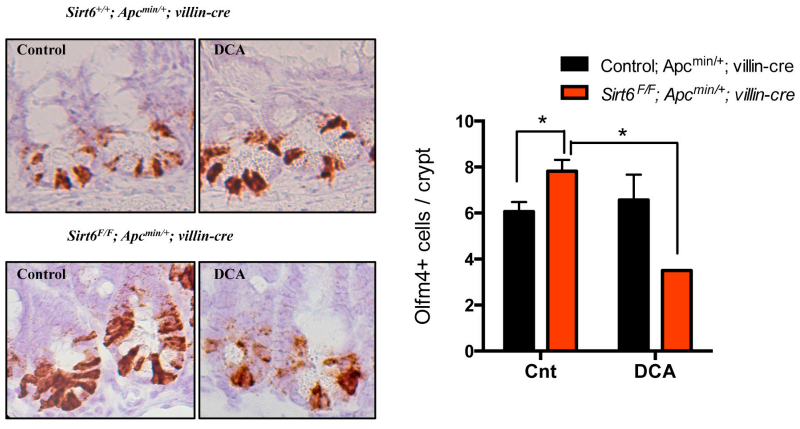

Figure 1. Expansion of ISCs in Sirt6$^{fl/fl}$; Villin-cre; Apc^{min} mice. Representative pictures of Olfm4 *in situ* hybridization showing ISC staining (left pannels) and quantification of Olfm4 postive cells (right pannel) in the intestines of control and Sirt6$^{fl/fl}$; Villin-cre; Apc^{min} mice treated or untreated with DCA.

has demonstrated that intestinal stem cells (ISCs) are the cell-of-origin of intestinal cancer in mice (Barker et al., 2009). Therefore, we hypothesized that SIRT6 could be regulating tumor initiation by increasing the number of ISCs, which, upon APC loss, could give rise to intestinal adenomas. To test this possibility, we started to analyze at the end of the first year whether lack of SIRT6 lead to an increase in the number and activity of ISCs. In preliminary studies, we found a 25% increase in the number of ISCs in the intestine of *Sirt6$^{fl/fl}$; villin-Cre; Apc^{min}* mice compared to control animals, a phenotype that was reversed by inhibiting glycolysis. We have now expanded these studies by analyzing more mice and confirmed that, indeed, loss of SIRT6 in the intestinal epithelium strikingly increases the number of ISCs in a glycolysis-dependent manner (Figure 1), suggesting that increased glycolysis in the absence of SIRT6 might be boosting the number of ISCs.

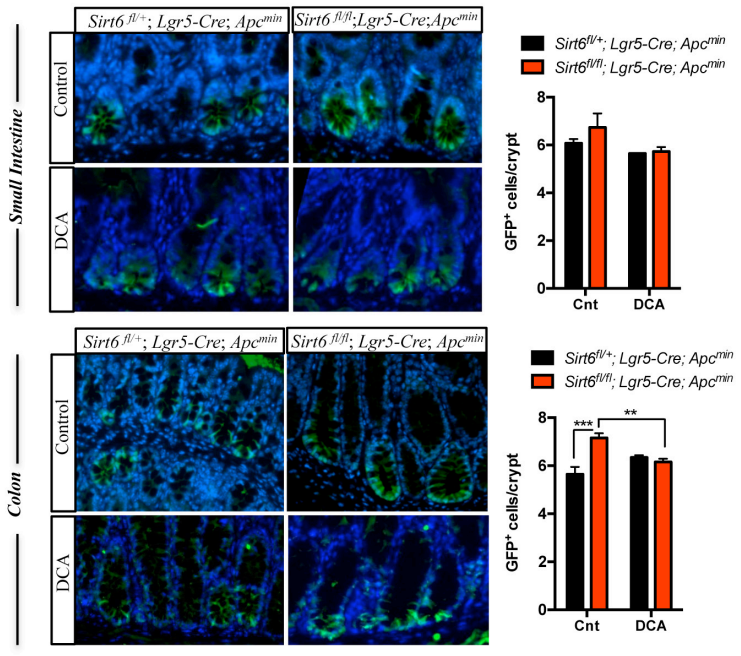

Figure 2. Expansion of Lgr5^{+} ISCs in Sirt6$^{fl/fl}$; Lgr5-EGFP-ires-CreERT2 mice. Representative pictures and quantification of Lgr5^{+} cells in small intestine and colon of control and Sirt6$^{fl/fl}$; Lgr5-EGFP-ires-CreERT2 mice. GFP expression was detected on intestine and colon paraffin sections by immunostaining with an anti-GFP atibody.

To corroborate that the effect of SIRT6 on ISC expansion and intestinal tumorigenesis is cell-autonomous, we planned to specifically delete SIRT6 on ISCs. To do so, we crossed our *Sirt6$^{fl/fl}$* mice with *Lgr5-EGFP-ires-CreERT2* mice (Barker et al., 2007). This system allows us to delete SIRT6 in Lgr5 positive cells (ISCs) as well as to visualize ISCs by GFP expression after tamoxifen injection. We have expanded our preliminary experiments and have found an increase in Lgr5 positive cells in both the intestine and colon of *Sirt6$^{fl/fl}$; Lgr5-EGFP-ires-CreERT2* mice, compared to control animals (Figure 2), suggesting that, indeed, deletion of SIRT6 in ISCs leads to an expansion of the ISC compartment, which represent putative tumor initiating cells. Next, we have moved to an *in vitro* system and derived intestinal organoids from isolated intestinal crypts from these mice. Strikingly, we have observed an increase in the number of organoids in *Sirt6$^{fl/fl}$; Lgr5-EGFP-ires-CreERT2* mice, indicating more ISC activity within crypts lacking SIRT6 (Figure 3). Again, this phenotype could be rescued by treating the mice with DCA, confirming our *in vivo* results (Figure 3).

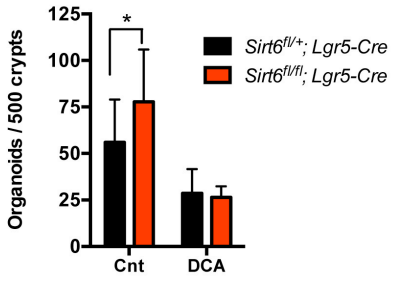

Figure 3. Increased organoid formation in the absence of SIRT6. Quantification of intestinal organoids derived from control and Sirt6$^{fl/fl}$; Lgr5-creERT2 mice untreated or treated with DCA.

All together, these results indicate that modulating glycolysis in ISCs can affect their activity. To better understand their metabolic requirements, we have isolated ISCs using the *Lgr5-EGFP-ires-CreERT2* mice described above. ISCs are defined as GFP^{high} cells, while progenitor cells and more differentiated epithelial cells express low levels of GFP (GFP^{low} cells) (Figure 4A and 4B). GFP^{high} and GFP^{low} cells were isolated by FACS (fluorescence activated cell sorting) and gene expression of ISC markers as well as glycolytic genes assessed by qPCR (Figure 4B). Remarkably, the expression of several key glycolytic genes (Glut1, Pfk1, Pdk1 and Ldhb) is upregulated in GFP^{high} cells, indicating that ISCs are activating a glycolytic gene expression program (Figure 4B). Furthermore, immunohistochemistry experiments on intestinal sections confirmed increased expression of PDK1 in the crypt compartment where ISCs are (Figure 4C).

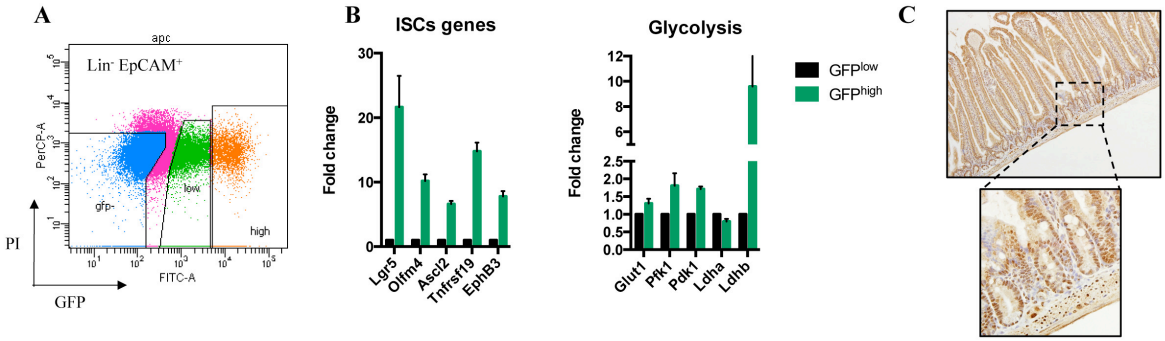

Figure 4. Increased glycolytic gene expression in ISCs. A) Cell sorting of GFP^{high} and GFP^{low} cells. B) Expression levels of ISC and glycolytic genes in sorted GFP^{high} and GFP^{low} cells. C) IHC showing high levels of PDK1 in the intestinal crypts.

To prove that high glycolytic metabolism is functionally relevant in ISCs, we analyzed organoid formation and growth by culturing freshly isolated crypts in the presence of DCA. As shown in figure 5, DCA treatment dramatically decreased the number and size of organoids compared to control conditions, highlighting the critical role of glucose metabolism reprogramming for ISCs function. Together, these results are in agreement with a model in which SIRT6 regulates ISCs activity by controlling glucose metabolism and, when lost, increased glycolysis promotes ISC expansion and the generation of potential tumor-initiating cells.

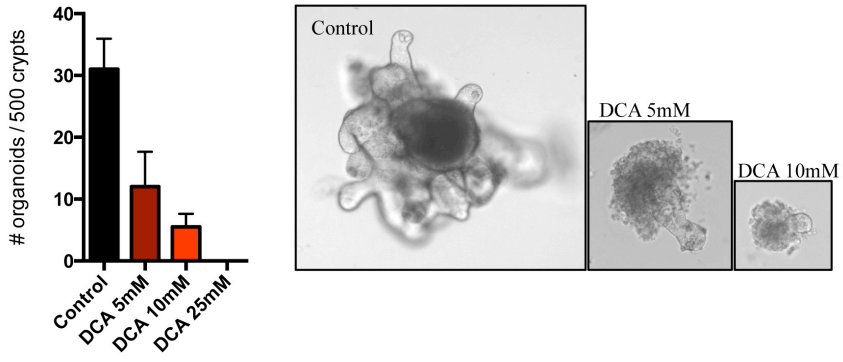

Figure 5. DCA inhibits ISCs activity. Number (left pannel) and representative pictures (right pannels) of intestinal organoids cultured in the absence or presence of DCA.

4. KEY RESEARCH ACCOMPLISHMENTS

- SIRT6 regulates the number of intestinal and adenoma stem cells by regulating glucose metabolism.
- ISCs are more glycolytic than more differentiated intestinal epithelial cells, and increased glycolysis in these cells regulates their stemness.

5. CONCLUSION

We have demonstrated that SIRT6 acts as a potent tumor suppressor in colorectal cancer *in vivo* by controlling glucose metabolic reprogramming. Importantly, inhibition of glycolysis in SIRT6-deficient mice dramatically reduces intestinal tumor initiation and growth, suggesting that targeting glycolysis might potentially provide a new approach to modulate cancer growth in those tumors with low SIRT6 levels. Data generated during this year has uncovered a role for SIRT6 as a critical regulator of ISC activity. Using two different mouse models as well as an *in vitro* intestinal organoid system, I have found that lack of SIRT6 increases the number and activity of ISCs, a phenotype that is reversed by inhibiting glycolysis, indicating that enhanced glycolytic metabolism in the absence of SIRT6 drives intestinal tumorigenesis by increasing the number of tumor-initiating cells. Importantly, an ISC signature identifies colorectal cancer stem cells in human patients and correlates with poorer disease outcome (Merlos-Suarez et al., 2011). Therefore, the identification of the precise mechanisms driving the genesis of these cells is of special relevance in order to design more effective therapeutic approaches. Lastly, we have described glucose metabolism requirements of ISCs that where previously unknown, which seems to be important in regulating the stemness of these cells. Further studies performed during next year will shed light on why ISCs need to have such a metabolic reprogramming and how aerobic glycolysis modulates their stemness, as well as its impact on intestinal regeneration and cancer.

6. PUBLICATIONS, ABSTRACTS AND PRESENTATIONS

Publications:

1. Lay press: nothing to report
2. Peer-reviewed scientific journals: nothing to report
3. Invited articles:
 - Sebastian C and Mostoslavsky R. Untangling the fiber yarn: butyrate feeds Warburg to suppress colorectal cancer. *Cancer Discov. 2014 Dec;4(12):1368-70.*
- Sebastian C and Mostoslavsky R. The role of mammalian sirtuins in cancer metabolism. *Semin Cell Dev Biol. 2015 Jul 31*
4. Abstracts: nothing to report

Presentations: SIRT6 suppresses intestinal tumorigenesis by regulating intestinal stem cells. Carlos Sebastian and Raul Mostoslavsky. Keystone Symposia: Biology of Sirtuins, Santa Fe, NM (March 8-12, 2015).

7. INVENTIONS, PATENTS AND LICENSES

Nothing to report

8. REPORTABLE OUTCOMES

Nothing to report

9. OTHER ACHIEVEMENTS

Nothing to report

10. REFERENCES

Barker, N., Ridgway, R. A., van Es, J. H., van de Wetering, M., Begthel, H., van den Born, M., Danenberg, E., Clarke, A. R., Sansom, O. J., and Clevers, H. (2009). Crypt stem cells as the cells-of-origin of intestinal cancer. Nature *457*, 608-611.

Barker, N., van Es, J. H., Kuipers, J., Kujala, P., van den Born, M., Cozijnsen, M., Haegebarth, A., Korving, J., Begthel, H., Peters, P. J., and Clevers, H. (2007). Identification of stem cells in small intestine and colon by marker gene Lgr5. Nature *449*, 1003-1007.

Merlos-Suarez, A., Barriga, F. M., Jung, P., Iglesias, M., Cespedes, M. V., Rossell, D., Sevillano, M., Hernando-Momblona, X., da Silva-Diz, V., Munoz, P.*, et al.* (2011). The intestinal stem cell signature identifies colorectal cancer stem cells and predicts disease relapse. Cell Stem Cell *8*, 511-524.

Mostoslavsky, R., Chua, K. F., Lombard, D. B., Pang, W. W., Fischer, M. R., Gellon, L., Liu, P., Mostoslavsky, G., Franco, S., Murphy, M. M.*, et al.* (2006). Genomic instability and aging-like phenotype in the absence of mammalian SIRT6. Cell *124*, 315-329.

Sebastian, C., Zwaans, B. M., Silberman, D. M., Gymrek, M., Goren, A., Zhong, L., Ram, O., Truelove, J., Guimaraes, A. R., Toiber, D.*, et al.* (2012). The histone deacetylase SIRT6 is a tumor suppressor that controls cancer metabolism. Cell *151*, 1185-1199.

Zhong, L., D'Urso, A., Toiber, D., Sebastian, C., Henry, R. E., Vadysirisack, D. D., Guimaraes, A., Marinelli, B., Wikstrom, J. D., Nir, T.*, et al.* (2010). The histone deacetylase Sirt6 regulates glucose homeostasis via Hif1alpha. Cell *140*, 280-293.

IN THE SPOTLIGHT

Untangling the Fiber Yarn: Butyrate Feeds Warburg to Suppress Colorectal Cancer

Carlos Sebastián and Raul Mostoslavsky

Summary: Dietary composition has an important role in shaping the gut microbiota. In turn, changes in the diet directly impinge on bacterial metabolites present in the intestinal lumen. Whether such metabolites play a role in intestinal cancer has been a topic of hot debate. In this issue of *Cancer Discovery*, Donohoe and colleagues show that dietary fiber protects against colorectal carcinoma in a microbiota-dependent manner. Furthermore, fiber-derived butyrate acts as a histone deacetylase inhibitor, inhibiting cell proliferation and inducing apoptosis in colorectal cancer cells experiencing the Warburg effect. *Cancer Discov; 4(12); 1368–70. ©2014 AACR.*

See related article by Donohoe and colleagues, p. 1387 (4).

Colorectal carcinoma is the third leading cause of cancer mortality in the world (1). This disease usually develops over many years via the accumulation of numerous genetic changes. Although some types of colorectal carcinoma are hereditary (2), most colorectal carcinoma cases are associated with diet and lifestyle (1). In line with this, the intestinal microbiota has been proposed to be a major contributor to the development of colorectal carcinoma (3). An increasing amount of data has demonstrated that dietary composition has an important effect on the gut microbiota, which, in turn, leads to changes in bacterial metabolites released to the intestinal lumen affecting intestinal tumorigenesis. In this context, dietary fiber is among the most-studied components of the diet in regard to the pathology of colorectal carcinoma. However, the role of fiber on colorectal carcinoma is controversial, mainly due to the fact that human cohort–based epidemiologic studies have yielded conflicting results. Furthermore, from those studies claiming a protective role, it is still unclear how fiber protects against colorectal carcinoma. Two possible mechanisms have been proposed. First, insoluble fiber may speed colonic transit, decreasing the exposure time of the colonic epithelium to carcinogens, and, second, intestinal bacteria can metabolize soluble fiber into metabolites with protective action, such as short-chain fatty acids (SCFA). In this issue, Donohoe and colleagues shed light on these controversies and elegantly demonstrate that, indeed, dietary fiber protects against colorectal carcinoma by increasing bacterial butyrate levels in the colon, which act as an histone deacetylase (HDAC) inhibitor, halting proliferation and promoting apoptosis of colon cancer cells (Fig. 1; ref. 4).

Genetic heterogeneity, differences in the composition of the gut microbiota, and the utilization of different sources of fiber are among the possible causes underlying the inconclusive results obtained from human studies (5). To overcome these hurdles, Donohoe and colleagues used BALB/c mice with a strictly defined gut microbiota kept on gnotobiotic isolators, thus avoiding colonization by other commensal bacteria (4). They then colonized some of the animals with *Butyrivibrio fibrosolvens*, a butyrate-producing bacterium, and fed the mice either low-fiber or high-fiber diets that were otherwise identical in composition and calorically matched. This experimental system allowed the authors to rule out any effect of differences in genetics, intestinal microbiota, and fiber source on colorectal carcinoma development. Using this gnotobiotic mouse model, they found that mice fed a high-fiber diet and colonized with *B. fibrosolvens* were protected against azoxymethane/dextran sodium sulfate (AOM/DSS)–induced colorectal carcinoma. Strikingly, these mice developed fewer, smaller, and less aggressive tumors than all the other experimental groups. Importantly, high-fiber diet *per se* did not have any protective effect on this colorectal carcinoma model, indicating that only in combination with the right microbiota could dietary fiber be beneficial in protecting against colorectal carcinoma. On the basis of these results, the authors propose that human epidemiologic studies should be revisited to incorporate differences in participants' gut microbiota to better address the role of dietary fiber on colorectal carcinoma.

Another important conclusion one can immediately draw from this result is that a metabolic product from fiber fermentation by *B. fibrosolvens* must be involved in the tumor-suppressive effect of dietary fiber. In line with this possibility, mice fed a high-fiber diet and colonized with *B. fibrosolvens* had increased luminal levels of butyrate, but not acetate and propionate, the other two major SCFAs. This result clearly points to butyrate as a key bacterial metabolite inhibiting colorectal carcinoma development. To confirm this hypothesis, the authors modulated luminal butyrate levels by two different means. First, they colonized mice with a mutant *B. fibrosolvens* strain (that produces 7-fold less butyrate when cultured) and fed them a

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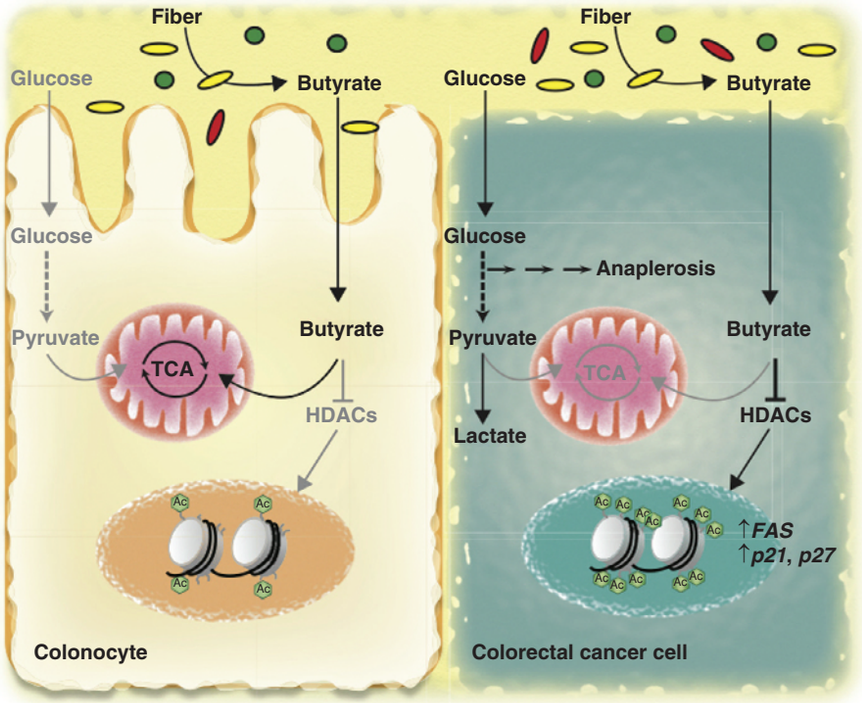

Figure 1. High-fiber diet leads to butyrate production in the colon by the action of butyrate-producing bacteria (yellow ovals), which is used by colonic epithelial cells as a primary source of energy. However, cancer cells use glucose to obtain energy and feed anaplerotic reactions (Warburg effect), leading to the accumulation of nonoxidized butyrate, which acts as an HDAC inhibitor, in turn increasing histone acetylation and expression of key proapoptotic and cell-cycle regulatory genes suppressing tumor growth.

low-fiber or high-fiber diet as before. After AOM/DSS treatment, they found that mutant *B. fibrosolvens* conferred an attenuated protective effect to high-fiber diet in these mice. Alternatively, they provided control mice a tributyrin-fortified diet, which increases colonic butyrate levels independently of microbiota. Following the AOM/DSS regimen, these mice were almost completely protected against colorectal carcinoma, indicating that exogenous butyrate could recapitulate the protective effect of high-fiber diet and *B. fibrosolvens*. Together, these two experiments clearly demonstrated that fiber fermentation by *B. fibrosolvens* protects from colorectal carcinoma by increasing luminal levels of bacterial butyrate.

The tumor-protective effect of butyrate has been mainly attributed to its anti-inflammatory properties. Butyrate downregulates the expression of proinflammatory cytokines in colonic macrophages, and it has been shown to regulate colonic regulatory T cells in mice, which have a crucial role in controlling intestinal inflammation (3). However, the authors did not find any difference in the number of regulatory T cells and associated cytokines among all the experimental groups, ruling out reduced inflammation as a cause for the protective effect of butyrate. On the basis of their previous work, the authors hypothesized that the tumor-suppressive role of butyrate in colorectal carcinoma could be related to the metabolic differences exhibited by normal and cancerous colonocytes. Butyrate represents the primary source of energy in normal colonic epithelial cells (6). However, colorectal carcinoma cells, like most cancer cells, display an increased glucose uptake and metabolism, a phenomenon termed the "Warburg effect" for the German scientist who originally described it in the early 20th century. Such a switch toward glycolytic metabolism is required to sustain their energetic and anaplerotic demands. As a consequence, butyrate is not catabolized in these cells to the same extent and, therefore, accumulates to such a concentration that it can act as an HDAC inhibitor (7). Indeed, the authors found increased levels of butyrate in the tumors of mice colonized with *B. fibrosolvens* and fed a high-fiber diet, suggesting that more butyrate molecules could be available to function as an HDAC inhibitor. Consistent with this, H3 acetylation levels are increased in the tumors of these mice, compared with adjacent normal colonocytes and tumors from control mice. Importantly, the authors found increased histone H3 acetylation at the promoter region of key proapoptotic and cell-cycle genes, such as *FAS*, *p21*, and *p27*, leading to increased expression of these genes and the concomitant inhibition of cell proliferation and induction of apoptosis of colorectal carcinoma cells. Finally, the authors extended these observations to human colorectal carcinoma samples, where they detected elevated levels of butyrate and H3 acetylation compared with matched normal mucosa.

Collectively, this body of work provides convincing evidence that dietary fiber, when combined with butyrate-producing bacteria, can protect from colorectal carcinoma by providing tumors with high levels of butyrate to act as an HDAC inhibitor, thus impairing tumor growth (Fig. 1). However, it also raises several intriguing questions. Which other species of bacteria are important in colorectal carcinoma protection? Although this study has focused on *B. fibrisolvens*, a type of bacteria common in ruminant animals, a large number of genera, including SCFA-producing species, have been identified in the human colon (8). In this context, different species could generate luminal butyrate at lower concentrations, inducing aberrant proliferation and transformation of colon epithelial cells, as recently reported in an $APC^{Min/+}MSH^{-/-}$ model of colorectal carcinoma (9). In the same way, can other bacterial metabolites play a role in

the tumor-suppressive effect of dietary fiber? The modest decrease in tumor protection shown in mice colonized with mutant *B. fibrisolvens* suggests that this very likely could be the case. It would be fascinating to elucidate which metabolites these are, and their effect on colorectal carcinoma prevention as well as in the metabolism of colon cancer cells. Finally, better knowledge of how to modulate our intestinal flora by changing our diet would definitely help us to elucidate the complex interaction between diet, intestinal flora, and colorectal carcinoma prevention.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. Cancer J Clin 2011;61:69–90.
2. Kinzler KW, Vogelstein B. Lessons from hereditary colorectal cancer. Cell 1996;87:159–70.
3. Louis P, Hold GL, Flint HJ. The gut microbiota, bacterial metabolites and colorectal cancer. Nat Rev Microbiol 2014;12:661–72.
4. Donohoe DR, Holley D, Collins LB, Montgomery SA, Whitmore AC, Hillhouse A, et al. A gnotobiotic mouse model demonstrates that dietary fiber protects against colorectal tumorigenesis in a microbiota- and butyrate-dependent manner. Cancer Discov 2014;4:1387–97.
5. Ferguson LR, Harris PJ. The dietary fibre debate: more food for thought. Lancet 2003;361:1487–8.
6. Roediger WE. Role of anaerobic bacteria in the metabolic welfare of the colonic mucosa in man. Gut 1980;21:793–8.
7. Donohoe DR, Collins LB, Wali A, Bigler R, Sun W, Bultman SJ. The Warburg effect dictates the mechanism of butyrate-mediated histone acetylation and cell proliferation. Mol Cell 2012;48:612–26.
8. Pryde SE, Duncan SH, Hold GL, Stewart CS, Flint HJ. The microbiology of butyrate formation in the human colon. FEMS Microbiol Lett 2002;217:133–9.
9. Belcheva A, Irrazabal T, Robertson SJ, Streutker C, Maughan H, Rubino S, et al. Gut microbial metabolism drives transformation of MSH2-deficient colon epithelial cells. Cell 2014;158:288–99.

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The role of mammalian sirtuins in cancer metabolism

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ABSTRACT

Metabolic reprogramming has recently emerged as a key feature of cancer cells, which need to rewire their cellular metabolism in order to sustain their faster proliferation and growth. New insight into the molecular mechanisms governing this metabolic reprogramming has implicated mammalian sirtuins as important regulators of cancer metabolism. Sirtuins are NAD^+-dependent protein deacylases involved in a variety of biological functions, including life span and health span regulation, genomic stability, tumorigenesis, inflammation, and metabolism. Due to the requirement of NAD^+ for their function, sirtuins can act as sensors of the metabolic state of the cell and regulate core metabolic pathways in response to cellular stresses, thus being good candidates to control the reprogramming of cellular metabolism that occurs during tumorigenesis. Here, we summarize our current knowledge of the roles of mammalian sirtuins in cancer metabolism, and discuss their implication in controlling this metabolic shift during aging and aging-associated cancers.

Contents

1. Introduction

One of the key features of cancer cells is their uncontrolled rate of cell division. In order to sustain this increased proliferation, cancer cells need to readjust their cellular metabolism to meet three basic requirements: ATP production to fulfill their energetic demands, biosynthesis of precursors to build up macromolecules, and maintenance of cellular redox status. Indeed, the observation that cancer cells display a different cellular metabolism than normal cells was first described by Otto Warburg more than 50 years ago [1]. In cancer cells, glucose-derived pyruvate is not fully oxidized in the TCA cycle. Instead, it is converted to lactate even in the presence of oxygen, and this metabolic switch is required for cancer cells to proliferate. This finding was interpreted for many years as an adaptive response of cancer cells to their increased proliferation. However, recent work has reset this idea by demonstrating that metabolic reprogramming is a fundamental trait of all cancer cells, one that is directly regulated by oncogenes and tumor suppressors [2,3]. Moreover, as described in other chapters of this series of reviews, extensive work done in the last decade has extended Warburg's observation to other core cellular metabolic pathways. In a similar way as glucose-derived carbon is shunted to biosynthetic reactions to build up amino acids, lipids and

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nucleotides, glutamine and serine metabolism are also rewired to provide carbon and nitrogen equivalents to sustain anaplerotic reactions [4,5]. Together, all these metabolic changes are essential to fuel cell proliferation and tumor growth and, therefore, cancer metabolism has been recently upgraded to a hallmark of cancer [6].

Several signaling pathways and transcription factors have been identified to regulate the rewiring of central metabolic pathways during tumorigenesis, such as activation of PI3K, stabilization of HIF-1, increased expression of MYC, suppression of the metabolic sensor AMPK and loss or mutation of the tumor suppressor p53 [2]. Importantly, new insight into the regulation of metabolic reprogramming in cancer cells has implicated the mammalian homologs of the yeast protein Sir2, the so-called sirtuins, as key molecular determinants in controlling cancer metabolism. In mammals, there are 7 sirtuin members (SIRT1–7), which play prominent roles in aging, metabolism, cancer, inflammation, DNA repair and cellular responses to stress [7]. Out of the seven sirtuins, SIRT3, SIRT4 and SIRT6 have been recently shown to directly regulate metabolic reprogramming in cancer cells. Moreover, SIRT1, SIRT2 and SIRT7 could potentially control cancer metabolism by modulating the activity of some important metabolic regulators. In this review, we summarize our current knowledge about the function of this family of proteins in regulating metabolism in the context of tumorigenesis and discuss their putative role in connecting cancer metabolism and aging.

2. Sirtuins

The founding member of the sirtuin family, Sir2 (silencing information regulator 2), was originally discovered as a silencing factor in *Saccharomyces cerevisiae* [8]. Almost two decades later, Sir2 came out as a top candidate in a screen for modulators of yeast life span [9]. Sir2 was found to promote longevity by suppressing the formation of extrachromosomal ribosomal DNA circles in yeast [10]. Soon after these findings, work done by the same laboratory demonstrated that Sir2 was a NAD^+-dependent deacetylase required for yeast life span extension upon calorie restriction (CR) [11,12]. The discovery of the biochemical activity of Sir2 and its implication in the aging process were the inception of a new field in biology that rapidly expanded: the study of Sir2 and its mammalian homologs, the sirtuins. Despite some controversy regarding Sir2 pro-longevity effects [13,14], these initial findings prompted many laboratories to characterize the function of the seven mammalian sirtuins, termed SIRT1–7.

All mammalian sirtuins contain a conserved catalytic domain including a large and structurally homologous Rossmann-fold domain for NAD^+ binding, and a more structurally diverse, smaller, zinc-binding domain [15]. However, their N- and C-termini differ in sequence and length, which might explain their different subcellular localization, targets and functions [7] (Fig. 1; Table 1). SIRT1 and SIRT2 are found in the cytoplasm and in the nucleus, SIRT3-5 are mitochondrial sirtuins while SIRT6 and SIRT7 are mainly found in the nucleus [16]. Biochemically, mammalian sirtuins are primarily NAD^+-dependent lysine deacetylases. However, some recent data has demonstrated that some sirtuins have evolved to remove other acyl groups, such as succinyl, malonyl, glutaryl and long chain fatty acyl groups [17–21]. Moreover, SIRT4 and SIRT6 possess ADP-ribosyltransferase activity, yet its biological relevance remains to be fully established [22–25]. Together, all these enzymatic activities are essential for mammalian sirtuins to modulate a variety of physiological processes, such as transcriptional regulation, genomic stability, cellular responses to stress, metabolism, inflammation, aging and cancer [7] (Fig. 1).

Mouse models to study the function of mammalian sirtuins have been recently generated. From the analysis of their genotype, one can immediately infer that this family of proteins has prominent roles in regulating metabolism. Due to their NAD^+ dependency, sirtuin activity is tightly linked to the metabolic state of the cell; thus, it has been proposed that sirtuins can act as metabolic sensors coordinating cellular responses to metabolic stresses. Indeed, nearly every sirtuin plays a role in regulating metabolism and energy homeostasis by controlling multiple metabolic pathways, such as lipid and glucose metabolism, ketone bodies synthesis, urea cycle and insulin secretion [7,26]. Furthermore, sirtuins appear to be key players in the beneficial effects of CR on healthy life span extension. CR promotes the expression of nicotinamide phosphoribosyltransferase (NAMPT), a rate-limiting enzyme of NAD^+ biosynthesis, leading to an increase in intracellular levels of NAD^+, and concomitantly, to the activation of sirtuin activity [11,27]. Importantly, SIRT1 and SIRT3 improve healthy aging in mice by protecting against several age-related pathologies [28–30], and brain-specific SIRT1- and whole body SIRT6-overexpressing mice exhibit extended life span [31,32], highlighting the importance of sirtuins in mammalian healthy life span and aging.

Aging is one of the most potent factors contributing to cancer, whose incidence increases exponentially with age [33]. Due to their role in regulating longevity and age-related pathologies, sirtuins represent good candidates to control tumorigenesis as well. Indeed, almost every sirtuin has been implicated in neoplastic transformation with both tumor suppressive and promoting functions depending on the specific tumor type, cellular context and signaling pathway affected [7,34]. Initially, most of the studies involving sirtuins in cancer focused on their ability to control cellular stress responses and DNA repair. However, as detailed below, a large body of evidence has recently shown that sirtuins have a prominent role in tumorigenesis by regulating energy metabolism in cancer cells [35–39]. Thus, it appears that this family of proteins could be at the crossroad of metabolism, cancer and aging.

3. Mitochondrial sirtuins: controlling tumor growth from the powerhouse

3.1. SIRT3 acts as a tumor suppressor by inhibiting glycolysis

As mentioned before, SIRT3, SIRT4 and SIRT5 reside in the mitochondria, where they control numerous aspects of mitochondrial metabolism. SIRT3 is the most studied and best characterized among mitochondrial sirtuins. It has a very robust deacetylase activity and represents the major regulator of the mitochondrial acetylome. SIRT3 KO mice and cells exhibit increased mitochondrial protein acetylation [40,41], and SIRT3 has been shown to control the acetylation of a vast number of mitochondrial proteins in response to CR and fasting [42,43]. By deacetylating and controlling the activity of its targets, SIRT3 regulates multiple metabolic pathways, some of which could have profound consequences for tumor growth [26]. Indeed, SIRT3 has been found to be a tumor suppressor, mainly by inhibiting mitochondrial ROS production through deacetylation and activation of SOD2, IDH2 and FoxO3a [29,37,44–47]. Loss of SIRT3 leads to an increase in ROS levels, which via stabilization of HIF1α, drives a metabolic reprogramming towards aerobic glycolysis promoting tumor growth [37,44]. Furthermore, SIRT3 regulates glycolytic metabolism by deacetylating and activating pyruvate dehydrogenase (PDH)-E1α [48,49] PDH is a key glycolytic enzyme that converts pyruvate into acetyl-CoA, which will be further oxidized in the TCA. Reduced PDH activity is a feature of cancer cells, mainly due to overexpression of pyruvate dehydrogenase kinases (PDK), which phosphorylate and inactivate PDH. SIRT3 deacetylates PDH-E1α at lysine 321 increasing its activity, and thus promoting pyruvate oxidation in the mitochondria. Lack of SIRT3 or expression of a mutant PDH-E1α mimicking a

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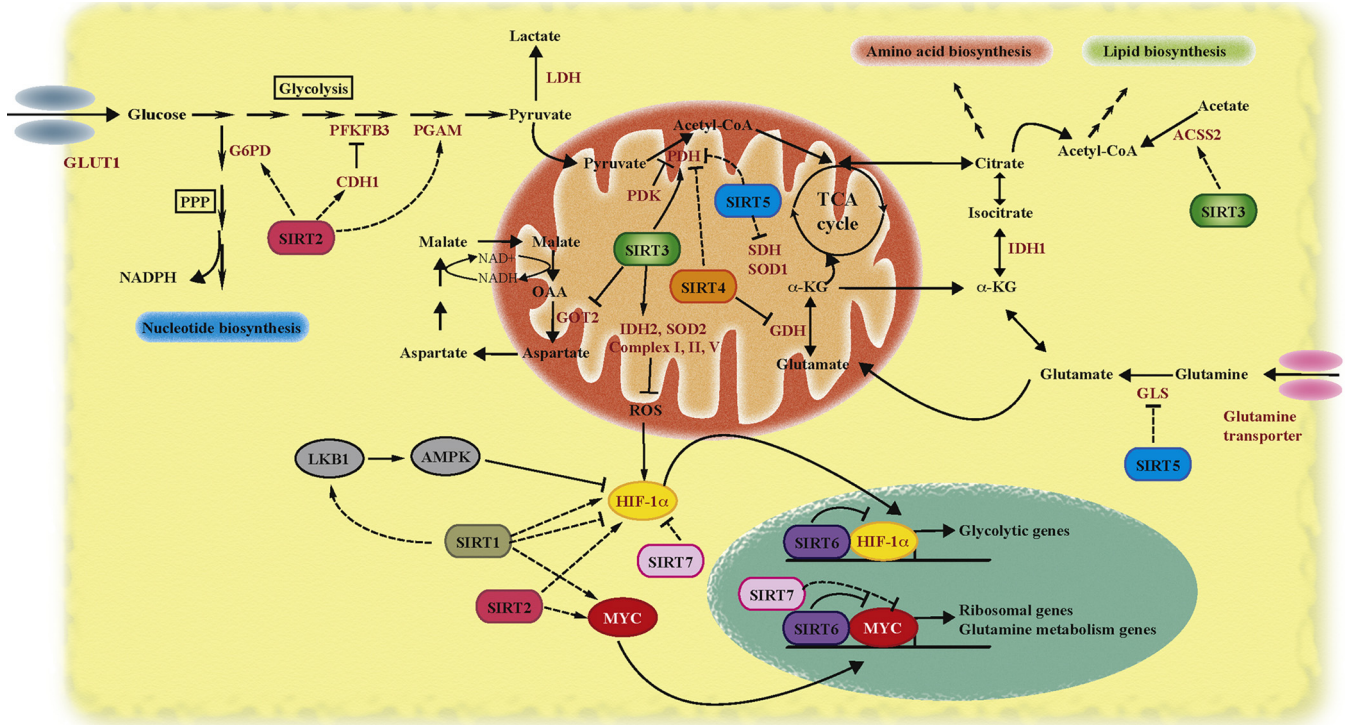

Fig. 1. Biological and metabolic functions of the different mammalian sirtuins. Dashed lines indicate known biological targets where their involvement in cancer remains to be established.

constitutively acetylated lysine 321 (K321Q) decrease PDH activity and promote aerobic glycolysis, increasing the tumorigenic potential of cancer cells [48]. Finally, SIRT3 has been recently described as a critical regulator of pancreatic tumor growth by controlling the malate-aspartate NADH shuttle activity [50]. This shuttle is important to transfer NADH from the cytosol into the mitochondria to sustain a high rate of glycolysis in tumor cells. SIRT3 deacetylates mitochondrial glutamate oxaloacetate transaminase (GOT2), a key enzyme controlling the malate-aspartate NADH shuttle, at lysines 159, 185 and 404. Increased GOT2 acetylation in absence of SIRT3 promotes the net transfer of cytosolic NADH into the mitochondria, supporting ATP production and cancer cell proliferation. Importantly, GOT2 K159 acetylation is increased in human pancreatic tumors, which correlates with reduced SIRT3 expression [50]. Moreover, SIRT3 expression is also downregulated in breast cancer, colon carcinoma, osteosarcoma, and hepatocellular carcinoma, and SIRT3 KO mice develop spontaneous mammary tumors, further supporting a tumor-suppressor role for this protein [37,44,45,51,52].

Despite not having been directly assessed, other SIRT3-regulated metabolic pathways could potentially have a role in metabolic rewiring during tumorigenesis. SIRT3 has a prominent

Table 1
Biochemical properties of mammalian sirtuins relevant in cancer metabolism.

Sirtuin	Cellular localization	Biochemical activity	Targets	Metabolic pathway
SIRT1	Cytoplasm	Deacetylase	HIF-1α/HIF-2α MYC	Unknown Glycolysis (*LDHa* expression)
	Nucleus	Deacetylase	Unknown	Unknown
SIRT2	Cytoplasm	Deacetylase	HIF-1α/CDH1/PGAM G6PD MYC	Glycolysis Pentose Phospate Pathway Unknown
	Nucleus	Deacetylase	Unknown	Unknown
SIRT3	Mitochondria	Deacetylase	SOD2/IDH2/FOXO3a PDH-E1α GOT2 Complex I, II, V	Glycolysis Glycolysis Mal-Asp NADH shuttle Unknown
	Cytoplasm	Deacetylase	ACSS2	Unknown
SIRT4	Mitochondria	ADP-ribosylase Lipoamidase	GDH PDH	Glutamine metabolism Unknown
SIRT5	Mitochondria	Desuccinylase	GLS PDH/SDH/SOD1	Unknown Unknown
SIRT6	Nucleus	Deacetylase	H3K9ac H3K56ac	Glycolysis Glutamine metabolism Ribosome biogenesis
SIRT7	Nucleus Cytoplasm	Deacetylase Deacetylase	H3K18ac HIF-1α/HIF-2α	Unknown Unknown

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role in promoting an efficient electron transport chain (ETC) function by deacetylating several components of the ETC complex I (such as NDUFA9), complex II (SDHA) and complex V (ATP synthase subunit β) [53–55]. In this way, SIRT3 maintains cellular ATP levels by promoting oxidative phosphorylation, thus avoiding potential activation of a Warburg-like metabolism. Moreover, keeping an efficient ETC reduces the levels of reactive oxygen species (ROS), which, as noted before, are important regulators of the Warburg effect. On the other hand, SIRT3 regulates acetate metabolism by deacetylating and activating acetyl-CoA synthetase 2 (ACSS2) [56,57]. ACSS2 catalyzes the conversion of acetate into acetyl-CoA and, as recently reported, it is overexpressed in cancer cells to produce enough acetyl-CoA to build up lipid biomass to fuel cell proliferation, in particular under hypoxic conditions [58–60]. In this context, it could be possible that SIRT3 could act as an oncogene to promote acetate utilization in cancer cells through activation of ACSS2. Indeed, it has been described that SIRT3 has some tumor promoting roles in specific types of cancer [61–63]. Overall, despite being described as a tumor suppressor, it is likely that SIRT3 may act as a tumor suppressor or an oncogene, in a tissue and context-specific manner.

3.2. SIRT4 limits tumor growth via repression of glutamine metabolism

Recent studies have shown that SIRT4 has important roles in cell metabolism and carcinogenesis as well. Despite having a conserved deacetylase domain, SIRT4 was initially described to have robust ADP-ribosyltransferase activity [22,24]. The mitochondrial enzyme glutamate dehydrogenase (GDH) was the first substrate of SIRT4 to be identified [24]. GDH is a key enzyme in glutamine metabolism and catalyzes the conversion of glutamate into α-ketoglutarate. SIRT4 ADP-ribosylates GDH to repress its activity resulting in a blockade of glutamine metabolism and a suppression of amino-acid-stimulated insulin secretion in pancreatic β-cells [24]. Recent data has demonstrated that SIRT4 also possesses a biologically relevant deacetylase activity. SIRT4 deacetylates and inhibits malonyl-CoA decarboxylase, the enzyme responsible for the generation of acetyl-CoA from malonyl-CoA, thus controlling fatty acid oxidation [64]. Moreover, SIRT4 also suppresses fatty acid oxidation by modulating the activity of PPARα and AMPK [65,66]. Finally, a new enzymatic activity has been recently associated with this mitochondrial sirtuin. SIRT4 can hydrolyze lipoamide groups from proteins, acting as a cellular lipoamidase [67]. In particular, SIRT4 removes lipoamide cofactors from the E2 component dihydrolipoyllysine acetyltransferase (DLAT) of PDH, diminishing its activity [67].

As observed for SIRT3, SIRT4 expression is downregulated in a variety of human tumors, including breast, colon, bladder, gastric, ovarian, thyroid and lung cancers, supporting a role for this sirtuin as a tumor suppressor [35,68–70]. In line with this, SIRT4 KO mice display increased incidence of solid tumors, especially lung tumors [35]. Mechanistically, SIRT4 suppresses tumorigenesis by inhibiting glutamine metabolism and promoting genomic stability [35,36,71]. As mentioned before, cancer cells avidly uptake glutamine to use it as an anaplerotic substrate to feed the TCA cycle. Glutamine-derived α-ketoglutarate is used to replenish the TCA cycle to support the biosynthesis of other amino acids and fatty acids. Moreover, in tumor cells with defective mitochondria or under hypoxia, the TCA cycle is inhibited, yet glutamine can still contribute to lipid biosynthesis by reductive carboxylation of α-ketoglutarate into citrate through a reverse enzymatic reaction of isocitrate dehydrogenase (IDH) [72,73]. Therefore, by inhibiting the activity of GDH, SIRT4 controls this key step in glutamine metabolism reprogramming in cancer cells, in turn promoting tumor growth. In this line of evidence, it has been shown that DNA damaging agents induce the expression of SIRT4 to repress glutamine metabolism and halt cell proliferation, allowing the cell to repair the damage. Accordingly, loss of SIRT4 results in increased glutamine-dependent proliferation and stress-induced genomic instability, promoting tumor growth [35]. SIRT4-dependent inhibition of glutamine metabolism has been also reported to suppress MYC-driven human Burkitt lymphoma [71]. Due to the aberrant activation of the transcription factor MYC, which regulates glutamine metabolism [74,75], Burkitt lymphoma cells exhibit increased glutamine uptake and metabolism. Overexpression of SIRT4 in these cells reduces glutamine utilization and glutamine-dependent proliferation, and sensitizes them to glucose depletion. Moreover, SIRT4 deletion in a mouse model of Burkitt lymphoma increases tumorigenesis and mortality by increasing glutamine metabolism [71]. Finally, mTORC1 activation, which has been linked to glutamine addiction in cancer cells [76], represses SIRT4 expression by promoting proteasome-mediated degradation of cAMP-responsive element binding 2 (CREB2), a *SIRT4* transcriptional regulator [36]. Similarly, C-terminal-binding protein (CtBP), a transcription factor controlling tumor initiation, progression and metastasis, has been found to have an essential role in promoting glutaminolysis by directly repressing the expression of SIRT4 in cancer cells [77].

SIRT4 could also act as an oncogene by promoting glucose metabolic reprogramming in cancer cells via inhibition of PDH. As noted before, SIRT4 functions as a lipoamidase to diminish PDH activity, a feature of cells experiencing Warburg effect. However, overexpression or increased activity of SIRT4 in tumors has not been documented to date. Therefore, further experiments are needed to confirm this hypothesis and determine the functional role of SIRT4 ADP-ribosyltransferase and lipoamidase activities in the context of tumorigenesis.

3.3. SIRT5 and cancer

Although initially considered a deacetylase based on its homology to other sirtuins [78], more recent work has demonstrated that SIRT5 functions primarily as a malonyl, succinyl and glutaryl deacylase [17,20,21,42,79]. As SIRT3 might be considered a regulator of the mitochondrial acetylome, SIRT5 has emerged as the sirtuin controlling the mitochondrial succinylome and glutarylome due to the large amount of substrates with significant increase in these post-translational modifications found in *SIRT5* KO mice and tissues. Among them, several proteins implicated in diverse metabolic pathways have been identified. SIRT5 regulates the urea cycle by deacetylating, desuccinylating and deglutarylating carbamoyl phosphate synthetase 1 (CPS1), in turn increasing its activity [17,21,80,81]. CPS1 catalyzes the first step of the urea cycle for ammonia detoxification in the liver and, to a lesser extent, in the kidney. Importantly, SIRT5 has also been reported to regulate ammonia production and ammonia-induced autophagy in non-liver cells by regulating glutamine metabolism. Mechanistically, SIRT5 desuccinylates and inhibits glutaminase, the enzyme catalyzing the conversion of glutamine into glutamate in a reaction producing ammonia [82]. Furthermore, SIRT5 plays an important role in ROS detoxification, as it desuccinylates and activates Cu/Zn superoxide dismutase (SOD1) [83], a key antioxidant enzyme. SIRT5 also controls glucose oxidation by repressing the activity of PDH-E1α and succinate dehydrogenase (SDH) by directly desuccinylating these enzymes [79]. Importantly, SOD1, PDH- E1α and SDH have been implicated in tumorigenesis. SOD1 expression has been found to be upregulated in breast cancer cell lines [84], and its overexpression correlates with increased growth of lung cancer cells [85,86]. Interestingly, mutation of the SOD1 succinylation site inhibits the growth of lung cancer cells, suggesting that SIRT5 could promote tumor growth by desuccinylating and activating SOD1 [83]. As noted before, suppression of PDH activity is important

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for cancer cells to reprogram their glucose metabolism. Moreover, inactivating mutations in SDH have been associated with tumorigenesis by stabilizing HIF-1α [87]. Therefore, SIRT5 could potentially act as an oncogene by inactivating these enzymes, thus promoting glucose metabolic reprogramming and tumor growth. In line with this, the genomic region comprising the *SIRT5* locus has been recently found to be amplified in 30% of human high-grade serous ovarian carcinomas, and SIRT5 is overexpressed in advanced non-small cell lung carcinoma [88,89].

4. SIRT6: bridging chromatin and cancer metabolism

SIRT6 is almost exclusively localized to chromatin, where it functions primarily as a histone deacetylase by removing acetyl groups from lysines 9 and 56 of histone H3 [90–92]. However, it has been reported that *in vitro* SIRT6 deacetylase activity is very weak, being about 1000 fold lower than SIRT1 deacetylase activity [18,19]. This discrepancy between SIRT6 *in vivo* and *in vitro* enzymatic activity has been in part attributed to the fact that SIRT6 requires to be in a full nucleosome context (rather that purified histones) to be fully active [93], and recent work demonstrated that SIRT6 deacetylase activity is highly increased by free fatty acids (which were not present in the initial *in vitro* studies) [18]. In addition to its deacetylase activity, SIRT6 also exhibits deacylase and ADP-ribosyltransferase activities. SIRT6 removes long-chain fatty acyl groups (myristoyl and palmitoyl) from lysine residues *in vitro* [18], and demyristoylates TNFα at lysines 19 and 20 allowing TNFα to be secreted by macrophages [19]. Although very weak, SIRT6 ADP-ribosyltransferase activity appears to be biologically relevant in the context of DNA damage upon oxidative stress, since SIRT6 ADP-risbosylates poly-ADP-ribose-polymerase (PARP1) stimulating its activity, thus promoting DNA repair [94]. Moreover, SIRT6-dependent ADP-ribosylation of the nuclear corepressor protein KAP1 contributes to the packaging of retrotransposons into transcriptionally repressive heterochromatin promoting genomic stability [95].

Through all its enzymatic activities, SIRT6 controls cellular homeostasis by regulating DNA repair [94,96–99], telomere maintenance [90], and glucose and lipid metabolism [100–104]. Importantly, the main phenotype that SIRT6 KO mice display is an acute and severe metabolic abnormality. Soon after birth, they develop a striking phenotype characterized by several acute degenerative processes, including loss of subcutaneous fat, lymphopenia, osteopenia, and acute onset of hypoglycemia, which lead to their death in less than 10 days [96]. This fatal drop in blood glucose levels is due to an increased glucose uptake by both muscle and brown adipose tissue. SIRT6-deficient cells avidly take up glucose, which is metabolized to lactate instead of being oxidized in the mitochondria [102]. Mechanistically, SIRT6 binds and co-represses HIF-1α transcriptional activity, thus suppressing the expression of several key glycolytic genes, such as glucose transporter-1 (*Glut1*), phosphofructokinase-1 (*Pfk1*), pyruvate dehydrogenase kinase-1 (*Pdk1*) and lactate dehydrogenase (*Ldh*) by specifically deacetylating H3K9 at the promoter region of these genes [102]. Accordingly, lack of SIRT6 leads to a glucose metabolic reprogramming that is reminiscent of the Warburg effect, suggesting that this sirtuin could suppress tumor growth. Indeed, SIRT6 functions as a tumor suppressor by repressing cancer metabolism. SIRT6 loss in immortalized mouse embryonic friboblasts (MEFs) leads to tumor formation even in the absence of any oncogene activation [38]. Metabolic reprogramming has been traditionally associated with the activation of oncogenic pathways, which, in addition to their effects on cell proliferation, directly impinge on core metabolic pathways. However, given the connection between these two processes, it has been difficult to discern cause from effect. In this context, these results represent one of the first evidence supporting a driving role for glucose metabolism reprogramming in tumorigenesis [105]. Importantly, inhibition of glucose metabolism by knocking-down PDK1 suppresses tumorigenesis in SIRT6-deficient cells, confirming the driver role of glycolytic metabolism in cancer initiation and growth [38]. In addition to promoting glycolysis, lack of SIRT6 leads to an increase in glutamine metabolism and ribosomal gene expression, although this phenotype appears to be a later event in the tumorigenic process. SIRT6 deacetylates H3K56 at the promoter region of these genes and co-represses MYC transcriptional activity, a master regulator of glutamine metabolism and ribosome biogenesis [38]. Furthermore, SIRT6 deletion in a mouse model of colorectal cancer increases the number, size and aggressiveness of adenomas, a phenotype that is fully suppressed by pharmacologically inhibiting PDK1 with dichloroacetate (DCA), highlighting the prominent role of glycolysis in SIRT6-dependent tumorigenesis [38]. Finally, SIRT6 expression is downregulated in human pancreatic, colorectal, liver and head and neck cancers [38,106–108]. Several mechanisms have been proposed to regulate SIRT6 levels in cancer cells, including proteasomal degradation in colon cancer cells and non-small cell lung cancer [109,110], and direct repression of its expression in breast, bladder and prostate cancer cells [111,112].

Like other sirtuins, increased SIRT6 expression has also been reported in few tumor types, such as pancreatic, prostate and breast cancers, where high SIRT6 levels are associated with chemotherapy resistance and poor prognosis [113–115]. In these tumors, the effect of SIRT6 on metabolism may not be dominant, but instead overexpression of SIRT6 may promote a pro-inflammatory phenotype or offer protection against chemotherapy-induced DNA damage, facilitating tumor growth.

5. SIRT1, SIRT2 and SIRT7: putative regulators of tumor metabolism

A large body of evidence has implicated SIRT1, SIRT2 and SIRT7 in neoplastic transformation. Although they have not been found to directly regulate cancer metabolism, recent data has uncovered new roles for these sirtuins in the regulation of key factors controlling metabolic reprogramming in cancer cells. As detailed below, SIRT1, SIRT2 and SIRT7 regulate the activity of both HIF and MYC proteins; SIRT1 also regulates the metabolic sensor liver kinase B1 (LKB1), and SIRT2 regulates de activity of several metabolic enzymes. Therefore, it is reasonable to speculate that these sirtuins could play important roles in tumorigenesis by regulating some aspects of tumor metabolism, a possibility that has not been fully explored yet.

SIRT1 was the first family member shown to function as a tumor suppressor, mainly by modulating cellular stress responses and DNA repair [116]. However, SIRT1 has also been found to promote tumor growth; therefore, like for other sirtuins, the role of SIRT1 in tumorigenesis seems to depend on the specific tumor type, cellular context and signaling pathway affected. SIRT1 possesses a very strong deacetylase activity and, accordingly, it regulates tumorigenesis by deacetylating and controlling the activity of a large variety of transcription factors, some of them key regulators of metabolic reprogramming in cancer cells. It was initially reported that SIRT1 was able to deacetylate and activate HIF-2α but not HIF-1α [117], although later studies confirmed a role for SIRT1 in regulating HIF-1α as well [118,119]. SIRT1 was found to deacetylate HIF-1α inhibiting its transcriptional activity by blocking HIF-1α-dependent recruitment of the p300 acetyltransferase to the promoters of HIF-1α target genes. In this setting, SIRT1 could repress HIF-1α-driven metabolic reprogramming [118]. However, these results have been challenged by another study, which showed

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a positive effect of SIRT1 on HIF-1α activity [119]. This study demonstrated that *in vivo* inhibition of SIRT1 activity in mice bearing hepatocellular carcinoma xenografts reduces tumor growth by downregulating the expression of *VEGFA* and blood vessel formation. In line with this, lack of SIRT1 in myeloid-derived suppressor cells (MDSC) promotes a HIF-1α-dependent glycolytic switch that skews these cells towards a pro-inflammatory phenotype, which confer protection against tumors [120]. Finally, SIRT1 can have differential roles in HIF-1α and HIF-2α mediated hypoxic responses depending on the cell type [121], adding more complexity to the functional interplay between SIRT1 and HIF proteins.

In addition to HIF, SIRT1 also cooperates with the oncogenic transcription factor MYC. MYC directly binds to the *SIRT1* promoter to induce its expression, which in turn deacetylates MYC at its C-terminal domain [122–124]. Although initially reported that SIRT1-mediated deacetylation of MYC was responsible for its degradation and, consequently, decreased MYC function [123], more recent studies have demonstrated that MYC deacetylation leads to its stabilization and increases MYC-MAX association and activity [122,124]. In this context, SIRT1 also forms a positive feedback loop with N-Myc to promote tumorigenesis in a mouse model of neuroblastoma, where N-Myc induces the expression of SIRT1, and SIRT1 stabilizes N-Myc protein by promoting MKP3-dependent phosphorylation of N-Myc [125]. Despite all these data supporting a role for SIRT1 in regulating MYC activity, it is currently unknown whether SIRT1 regulates MYC-dependent metabolic reprogramming in cancer cells. In this regard, it has been recently shown that SIRT1 stimulates MYC-induced *LDHA* expression [122,126], suggesting that, indeed, SIRT1 could regulate some of the metabolic effects of MYC in cancer.

Finally, it has been reported that SIRT1 deacetylates and activates LKB1 [127], a known tumor suppressor that regulates several metabolic pathways. One of the main targets for LKB1 is AMP-activated protein kinase (AMPK), a key enzyme that regulates cellular energy homeostasis [128]. LKB1 phosphorylates AMPK at Thr-172, an event required for its activation [129]. Importantly, AMPK has been recently described to be a tumor suppressor by negatively regulating the Warburg effect [130], raising the possibility that SIRT1 could suppress metabolic reprogramming in cancer cells by activating the LKB1-AMPK axis.

SIRT2 also functions as a tumor suppressor, mainly by promoting genomic stability. SIRT2 regulates mitotic progression by controlling the activity of the anaphase promoting complex/cyclosome via deacetylation of its coactivators CDH1 and CDC20, and thus prevents chromosomal instability during mitosis [131]. Furthermore, SIRT2 binds and deacetylates the methyltransferase PR-Set7, modulating its chromatin localization and resulting in H4K20 methylation, a chromatin mark involved in genomic stability [132]. Interestingly, CDH1 has been shown to inhibit glycolysis and cancer cell proliferation through ubiquitination and degradation of the glycolytic enzyme 6-phosphofructo-2-kinase/fructose-2,6-bisphosphatase, isoform 3 (PFKFB3) [133], suggesting that SIRT2-mediated deacetylation of CDH1 could regulate aerobic glycolysis. Supporting this hypothesis, SIRT2 has been recently found to regulate the activity of HIF-1α by deacetylating lysine 709 and promoting its stability. Importantly, HIF-1α induction by SIRT2 deficiency increases the expression of the glycolytic genes *GLUT1* and *LDHA*, supporting tumor growth [134]. Furthermore, SIRT2 also regulates the activity of phosphoglycerate mutase (PGAM) and glucose-6-phosphate dehydrogenase (G6PD), two key metabolic enzymes involved in glycolysis and the pentose phosphate pathway (PPP). PGAM is a glycolytic enzyme that catalyzes the conversion of 3-phosphoglycerate (3-PG) to 2-phosphoglycerate (2-PG). By controlling intracellular levels of 3-PG and 2-PG, PGAM coordinates glycolysis and biosynthesis to promote tumor growth [135]. Xu and colleagues found that PGAM activity is regulated by acetylation at lysine 100, and SIRT2 deacetylates this residue to activate its enzymatic activity [136]. Although SIRT2-dependent deacetylation of PGAM was not confirmed to be relevant for tumor growth *in vivo*, overexpression of a PGAM acetylation mimetic mutant reduced cell proliferation and tumor growth [136], highlighting the importance of this acetylation in controlling PGAM activity in the context of tumorigenesis. As noted before, SIRT2 also deacetylates G6PD, which catalyzes the conversion of glucose-6-phosphate to 6-phosphogluconate generating NADPH, and represents the rate-limiting and primary control step of the NADPH-generating branch in the PPP. SIRT2-mediated deacetylation of G6PD activates its enzymatic activity and stimulates PPP to supply cytosolic NADPH promoting cell survival during oxidative stress [137]. Since an active PPP is important in tumor cells for nucleotide biosynthesis and redox homeostasis, this result imply that SIRT2 could potentially promote tumor growth by activating this metabolic pathway.

As for SIRT1, the MYC oncoproteins also form a positive feedback loop with SIRT2. Both MYC and N-MYC induce the expression of SIRT2, which in turn deacetylates and stabilizes them, promoting growth of neuroblastoma and pancreatic cancer cells [138]. However, whether the interplay between SIRT2 and MYC proteins is relevant in the context of cancer metabolism remains unexplored.

Lastly, although the role of SIRT7 in tumorigenesis has been less studied, a recent report showed pro-oncogenic properties for this sirtuin by deacetylating H3K18 and repressing transcription of multiple genes involved in anchorage-independent growth and contact inhibition [139]. In line with this, SIRT7 has been also described to act as an oncogene in hepatocellular carcinoma, gastric cancer and colorectal cancer [140–142]. As other sirtuins, SIRT7 also regulates HIF proteins, potentially underlying the Warburg effect. SIRT7 binds and decreases the stability of both HIF-1α and HIF-2α, although the molecular mechanism involved in this regulation is not known [143]. Finally, SIRT7 also acts as a co-repressor of MYC to suppress ribosomal gene expression in response to endoplasmic reticulum (ER) stress [144]. Nevertheless, whether SIRT7 plays a role in cancer-associated metabolic reprogramming remains to be elucidated.

6. Sirtuins as modulators of cancer metabolism during aging

Medical advances and global heath policies have resulted in an increase in life expectancy. This shift in the distribution of population towards older ages has profound medical implications. Aging is accompanied with a major incidence of many diseases, and roughly two thirds of people who die each day in the world die of age-related causes (this percentage reaches 90% in industrialized countries) [145]. Remarkably, cancer ranks at the top of the list of age-related deaths and its incidence increases exponentially with age [33]. The most common explanation for the higher incidence of cancer during aging is the accumulation of mutations over time as we age, providing aging enough time for these mutations to reach the threshold required for tumor initiation and growth. However, this theory has some caveats as it fails to explain why some metabolic diseases, such as type II diabetes or obesity, exhibit a tight correlation with cancer propensity and worse prognosis [146]. In line with this, metabolic alterations via CR or physical exercise drastically reduce the risk of developing cancer [147]. These observations suggest that a decrease in metabolic homeostasis as we age could be an important factor contributing to the increased incidence of cancer during aging. In this setting, metabolism in tissues is known to change with age, with a clear decrease in mitochondrial respiration coupled to an increased in lactate metabolism [148,149]. Moreover, some tissues

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exhibit increased aerobic glycolysis with age, mainly due to upregulated activities of several glycolytic enzymes [150–152]. These metabolic changes are remarkably similar to cancer metabolism reprogramming, raising the possibility that age-associated changes in metabolism could predispose to tumorigenesis and promote tumor growth.

Increased ROS production, p53 and NF-κB have been involved in this age-dependent metabolic switch [153]. However, the precise molecular players directly regulating this adaptation remain poorly understood. As mentioned before, sirtuins have emerged as key regulators of metabolic rewiring during tumorigenesis, and they play a crucial role in regulating healthy lifespan and aging. Therefore, they represent excellent candidates to connect cancer, aging and metabolic reprogramming. Although there is no data directly linking sirtuins as regulators of age-associated cancers by controlling metabolism, several lines of evidence support this hypothesis. The levels of NAD^+ decrease in multiple tissues during aging, mainly due to a downregulation of nicotinamide phosphoribosyltransferase (NAMPT), the rate-limiting enzyme in mammalian NAD^+ biosynthesis [154]. Since sirtuins require NAD^+ for their activity, this result suggests that the function of these proteins could be impaired during aging. Importantly, nicotinamide mononucleotide (NMN), a product of the NAMPT reaction and a key NAD^+ intermediate, ameliorates the metabolic defects associated with decreased NAD^+ during aging, partly through SIRT1 activation [154]. In agreement with this, SIRT1 overexpression in mice improves healthy aging, while decreasing the incidence of spontaneous carcinomas and sarcomas. These effects, however, are not sufficiently potent to affect longevity, but clearly suggest a protective role for SIRT1 in age-associated cancers [28]. Furthermore, CR induces the expression of SIRT1 in several tissues [155], and overexpression or pharmacological activation of SIRT1 mimics the protective effects of CR on age-associated metabolic decline, neurodegeneration and cancer [156]. Remarkably, SIRT1 has been recently involved in the establishment of a Warburg-like phenotype during aging by stabilizing HIF-1α and disrupting mitochondrial homeostasis [157]. Drop of NAD^+ levels occurs during aging, in turn decreasing SIRT1 activity, causing VHL levels to decline promoting the stabilization of HIF-1α. The activation of this program induces a pseudohypoxic state that disrupts nuclear-mitochondrial communication and OXPHOS, which could promote tumorigenesis by increasing ROS levels and mutagenesis [157]. Importantly, a decline in mitochondrial function with age has been linked to loss of cellular homeostasis and organismal health [158]. Besides SIRT1, mitochondrial sirtuins also regulate mitochondrial metabolism as well as some aspects of cancer metabolism and, thus, they could also control age-associated cancers by impacting on mitochondrial homeostasis. Furthermore, SIRT3 and SIRT6 also regulate HIF-1α activity, raising the possibility that these sirtuins could contribute as well to the acquisition of this pseudohypoxic state during aging. Finally, SIRT6 overexpression extends lifespan in male mice [32], a phenotype that has been partially attributed to its role as a tumor suppressor [159]. Due to the driving role of SIRT6 in tumorigenesis by controlling glucose metabolic reprogramming [38], it is tempting to speculate that loss of SIRT6 (levels or activity) could contribute to the Warburg-like metabolic shift observed during aging, promoting tumorigenesis.

7. Concluding remarks

Extensive work done over the past few years has greatly improved our understanding of the biology behind mammalian sirtuins, which have been involved in a steadily growing list of physiological processes. Due to their NAD^+-dependent activity, sirtuins function as cellular rheostats controlling core metabolic pathways and, thus, are essential in maintaining metabolic homeostasis. Changes in carbon and nitrogen metabolism have been shown to be strictly required for tumor initiation and progression and, accordingly, cancer cells need to reprogram their metabolism to fulfill the energetic and anabolic demands of cell proliferation. Not surprisingly, as metabolic sensors, several sirtuins have been uncovered as central regulators of this metabolic reprogramming by modulating mitochondrial metabolism (SIRT3 and SIRT4) and epigenetics (SIRT6). Although the role of other sirtuins in driving cancer metabolism has not been explored yet, they clearly regulate some of the metabolic pathways altered in cancer and, with no doubt, future work will likely involve these sirtuins in tumorigenesis by controlling metabolism in cancer cells. However, further work seems necessary to understand how sirtuins' expression/activity is regulated during tumorigenesis, and whether their roles in cancer metabolism are general or specific for some types of tumors. Moreover, almost every sirtuin has other relevant functions in cancer cells, from promoting DNA repair to controlling the activity of transcription factors, and depending on the dominant role of some of these activities, sirtuins will have tumor promoting or suppressing roles. Therefore, elucidating the precise interplay of these functions in a particular tumor will be essential to design drugs to efficiently target this family of proteins. In this regard, controlling the activity of sirtuins could have an enormous impact on age-related diseases, including cancer. As discussed before, the role of sirtuins as regulators of cancer metabolism could be one of the mechanisms by which they promote longevity and health span, defining this family of proteins as key modulators at the intersection of cancer, metabolism and aging.

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References

[1] Warburg O. On the origin of cancer cells. Science 1956;123:309–14.
[2] Cairns RA, Harris IS, Mak TW. Regulation of cancer cell metabolism. Nat Rev Cancer 2011;11:85–95.
[3] Lunt SY, Vander Heiden MG. Aerobic glycolysis: meeting the metabolic requirements of cell proliferation. Annu Rev Cell Dev Biol 2011;27:441–64.
[4] Hensley CT, Wasti AT, DeBerardinis RJ. Glutamine and cancer: cell biology, physiology, and clinical opportunities. J Clin Invest 2013;123:3678–84.
[5] Locasale JW. Serine, glycine and one-carbon units: cancer metabolism in full circle. Nat Rev Cancer 2013;13:572–83.
[6] Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011;144:646–74.
[7] Sebastian C, Satterstrom FK, Haigis MC, Mostoslavsky R. From sirtuin biology to human diseases: an update. J Biol Chem 2012;287:42444–52.
[8] Klar AJ, Strathern JN, Broach JR, Hicks JB. Regulation of transcription in expressed and unexpressed mating type cassettes of yeast. Nature 1981;289:239–44.
[9] Kaeberlein M, McVey M, Guarente L. The SIR2/3/4 complex and SIR2 alone promote longevity in *Saccharomyces cerevisiae* by two different mechanisms. Genes Dev 1999;13:2570–80.
[10] Sinclair DA, Guarente L. Extrachromosomal rDNA circles—a cause of aging in yeast. Cell 1997;91:1033–42.
[11] Imai S, Armstrong CM, Kaeberlein M, Guarente L. Transcriptional silencing and longevity protein Sir2 is an NAD- dependent histone deacetylase. Nature 2000;403:795–800.
[12] Lin SJ, Defossez PA, Guarente L. Requirement of NAD and SIR2 for life-span extension by calorie restriction in *Saccharomyces cerevisiae*. Science 2000;289:2126–8.
[13] Burnett C, Valentini S, Cabreiro F, Goss M, Somogyvari M, Piper MD, et al. Absence of effects of Sir2 overexpression on lifespan in *C. elegans* and Drosophila. Nature 2011;477:482–5.
[14] Longo VD, Kennedy BK. Sirtuins in aging and age-related disease. Cell 2006;126:257–68.

Please cite this article in press as: Sebastián C, Mostoslavsky R. The role of mammalian sirtuins in cancer metabolism. Semin Cell Dev Biol (2015), http://dx.doi.org/10.1016/j.semcdb.2015.07.008

[15] Feldman JL, Dittenhafer-Reed KE, Denu JM. Sirtuin catalysis and regulation. J Biol Chem 2012;287:42419–27.
[16] Michishita E, Park JY, Burneskis JM, Barrett JC, Horikawa I. Evolutionarily conserved and nonconserved cellular localizations and functions of human SIRT proteins. Mol Biol Cell 2005.
[17] Du J, Zhou Y, Su X, Yu JJ, Khan S, Jiang H, et al. Sirt5 is a NAD-dependent protein lysine demalonylase and desuccinylase. Science 2011;334:806–9.
[18] 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;288:31350–6.
[19] Jiang H, Khan S, Wang Y, Charron G, He B, Sebastian C, et al. SIRT6 regulates TNF-alpha secretion through hydrolysis of long-chain fatty acyl lysine. Nature 2013;496:110–3.
[20] C. Peng, Z. Lu, Z. Xie, Z. Cheng, Y. Chen, M. Tan, et al., The first identification of lysine malonylation substrates and its regulatory enzyme, Mo. Cell. Proteomics 10 (2011) M111 012658.
[21] Tan M, Peng C, Anderson KA, Chhoy P, Xie Z, Dai L, et al. Lysine glutarylation is a protein posttranslational modification regulated by SIRT5. Cell Metab 2014;19:605–17.
[22] Ahuja N, Schwer B, Carobbio S, Waltregny D, North BJ, Castronovo V, et al. Regulation of insulin secretion by SIRT4, a mitochondrial ADP-ribosyltransferase. J Biol Chem 2007;282:33583–92.
[23] Frye RA. Characterization of five human cDNAs with homology to the yeast SIR2 gene: Sir2-like proteins (sirtuins) metabolize NAD and may have protein ADP-ribosyltransferase activity. Biochem Biophys Res Commun 1999;260:273–9.
[24] Haigis MC, Mostoslavsky R, Haigis KM, Fahie K, Christodoulou DC, Murphy AJ, et al. SIRT4 inhibits glutamate dehydrogenase and opposes the effects of calorie restriction in pancreatic beta cells. Cell 2006;126:941–54.
[25] Liszt G, Ford E, Kurtev M, Guarente L. Mouse Sir2 homolog SIRT6 is a nuclear ADP-ribosyltransferase. J Biol Chem 2005;280:21313–20.
[26] Kumar S, Lombard DB. Mitochondrial sirtuins and their relationships with metabolic disease and cancer. Antioxid Redox Signal 2015;22:1060–77.
[27] Houtkooper RH, Canto C, Wanders RJ, Auwerx J. The secret life of NAD+: an old metabolite controlling new metabolic signaling pathways. Endocr Rev 2010;31:194–223.
[28] Herranz D, Munoz-Martin M, Canamero M, Mulero F, Martinez-Pastor B, Fernandez-Capetillo O, et al. Sirt1 improves healthy ageing and protects from metabolic syndrome-associated cancer. Nat Commun 2010;1:3.
[29] Someya S, Yu W, Hallows WC, Xu J, Vann JM, Leeuwenburgh C, et al. Sirt3 mediates reduction of oxidative damage and prevention of age-related hearing loss under caloric restriction. Cell 2010;143:802–12.
[30] Guarente L, Franklin H, Epstein Lecture:. Sirtuins, aging, and medicine. N Engl J Med 2011;364:2235–44.
[31] Satoh A, Brace CS, Rensing N, Cliften P, Wozniak DF, Herzog ED, et al. Sirt1 extends life span and delays aging in mice through the regulation of Nk2 homeobox 1 in the DMH and LH. Cell Metab 2013;18:416–30.
[32] Kanfi Y, Naiman S, Amir G, Peshti V, Zinman G, Nahum L, et al. The sirtuin SIRT6 regulates lifespan in male mice. Nature 2012;483:218–21.
[33] Jemal A, Siegel R, Xu J, Ward E. Cancer statistics, 2010. CA: Cancer J Clin 2010;60:277–300.
[34] Roth M, Chen WY. Sorting out functions of sirtuins in cancer. Oncogene 2014;33:1609–20.
[35] Jeong SM, Xiao C, Finley LW, Lahusen T, Souza AL, Pierce K, et al. SIRT4 has tumor-suppressive activity and regulates the cellular metabolic response to DNA damage by inhibiting mitochondrial glutamine metabolism. Cancer Cell 2013;23:450–63.
[36] Csibi A, Fendt SM, Li C, Poulogiannis G, Choo AY, Chapski DJ, et al. The mTORC1 pathway stimulates glutamine metabolism and cell proliferation by repressing SIRT4. Cell 2013;153:840–54.
[37] Finley LW, Carracedo A, Lee J, Souza A, Egia A, Zhang J, et al. SIRT3 opposes reprogramming of cancer cell metabolism through HIF1alpha destabilization. Cancer Cell 2011;19:416–28.
[38] Sebastian C, Zwaans BM, Silberman DM, Gymrek M, Goren A, Zhong L, et al. The histone deacetylase SIRT6 is a tumor suppressor that controls cancer metabolism. Cell 2012;151:1185–99.
[39] Guarente L. The many faces of sirtuins: Sirtuins and the Warburg effect. Nat Med 2014;20:24–5.
[40] Lombard DB, Alt FW, Cheng HL, Bunkenborg J, Streeper RS, Mostoslavsky R, et al. Mammalian Sir2 homolog SIRT3 regulates global mitochondrial lysine acetylation. Mol Cell Biol 2007;27:8807–14.
[41] Sol EM, Wagner SA, Weinert BT, Kumar A, Kim HS, Deng CX, et al. Proteomic investigations of lysine acetylation identify diverse substrates of mitochondrial deacetylase sirt3. PLoS ONE 2012;7:e50545.
[42] Rardin MJ, Newman JC, Held JM, Cusack MP, Sorensen DJ, Li B, et al. Label-free quantitative proteomics of the lysine acetylome in mitochondria identifies substrates of SIRT3 in metabolic pathways. Proc Natl Acad Sci USA 2013;110:6601–6.
[43] Hebert AS, Dittenhafer-Reed KE, Yu W, Bailey DJ, Selen ES, Boersma MD, et al. Calorie restriction and SIRT3 trigger global reprogramming of the mitochondrial protein acetylome. Mol Cell 2013;49:186–99.
[44] Bell EL, Emerling BM, Ricoult SJ, Guarente L. SirT3 suppresses hypoxia inducible factor 1alpha and tumor growth by inhibiting mitochondrial ROS production. Oncogene 2011;30:2986–96.
[45] Kim HS, Patel K, Muldoon-Jacobs K, Bisht KS, Aykin-Burns N, Pennington JD, et al. SIRT3 is a mitochondria-localized tumor suppressor required for maintenance of mitochondrial integrity and metabolism during stress. Cancer Cell 2010;17:41–52.
[46] Tao R, Coleman MC, Pennington JD, Ozden O, Park SH, Jiang H, et al. Sirt3-mediated deacetylation of evolutionarily conserved lysine 122 regulates MnSOD activity in response to stress. Mol Cell 2010;40:893–904.
[47] Sundaresan NR, Samant SA, Pillai VB, Rajamohan SB, Gupta MP. SIRT3 is a stress-responsive deacetylase in cardiomyocytes that protects cells from stress-mediated cell death by deacetylation of Ku70. Mol Cell Biol 2008;28:6384–401.
[48] Ozden O, Park SH, Wagner BA, Yong Song H, Zhu Y, Vassilopoulos A, et al. SIRT3 deacetylates and increases pyruvate dehydrogenase activity in cancer cells. Free Radic Biol Med 2014;76:163–72.
[49] Fan J, Shan C, Kang HB, Elf S, Xie J, Tucker M, et al. Tyr phosphorylation of PDP1 toggles recruitment between ACAT1 and SIRT3 to regulate the pyruvate dehydrogenase complex. Mol Cell 2014;53:534–48.
[50] Yang H, Zhou L, Shi Q, Zhao Y, Lin H, Zhang M, et al. SIRT3-dependent GOT2 acetylation status affects the malate-aspartate NADH shuttle activity and pancreatic tumor growth. EMBO J 2015;34:1110–25.
[51] Zhang CZ, Liu L, Cai M, Pan Y, Fu J, Cao Y, et al. Low SIRT3 expression correlates with poor differentiation and unfavorable prognosis in primary hepatocellular carcinoma. PLoS ONE 2012;7:e51703.
[52] Zhang YY, Zhou LM. Sirt3 inhibits hepatocellular carcinoma cell growth through reducing Mdm2-mediated p53 degradation. Biochem Biophys Res Commun 2012;423:26–31.
[53] Ahn BH, Kim HS, Song S, Lee IH, Liu J, Vassilopoulos A, et al. A role for the mitochondrial deacetylase Sirt3 in regulating energy homeostasis. Proc Natl Acad Sci USA 2008;105:14447–52.
[54] Cimen H, Han MJ, Yang Y, Tong Q, Koc H, Koc EC. Regulation of succinate dehydrogenase activity by SIRT3 in mammalian mitochondria. Biochemistry 2010;49:304–11.
[55] Rahman M, Nirala NK, Singh A, Zhu LJ, Taguchi K, Bamba T, et al. Drosophila Sirt2/mammalian SIRT3 deacetylates ATP synthase beta and regulates complex V activity. J Cell Biol 2014;206:289–305.
[56] Hallows WC, Lee S, Denu JM. Sirtuins deacetylate and activate mammalian acetyl-CoA synthetases. Proc Natl Acad Sci USA 2006;103:10230–5.
[57] Schwer B, Bunkenborg J, Verdin RO, Andersen JS, Verdin E. Reversible lysine acetylation controls the activity of the mitochondrial enzyme acetyl-CoA synthetase 2. Proc Natl Acad Sci USA 2006;103(27):10224–9.
[58] Comerford SA, Huang Z, Du X, Wang Y, Cai L, Witkiewicz AK, et al. Acetate dependence of tumors. Cell 2014;159:1591–602.
[59] Mashimo T, Pichumani K, Vemireddy V, Hatanpaa KJ, Singh DK, Sirasanagandla S, et al. Acetate is a bioenergetic substrate for human glioblastoma and brain metastases. Cell 2014;159:1603–14.
[60] Schug ZT, Peck B, Jones DT, Zhang Q, Grosskurth S, Alam IS, et al. Acetyl-CoA synthetase 2 promotes acetate utilization and maintains cancer cell growth under metabolic stress. Cancer Cell 2015;27:57–71.
[61] Alhazzazi TY, Kamarajan P, Joo N, Huang JY, Verdin E, D'Silva NJ, et al. Sirtuin-3 (SIRT3), a novel potential therapeutic target for oral cancer. Cancer 2011;117:1670–8.
[62] Ashraf N, Zino S, Macintyre A, Kingsmore D, Payne AP, George WD, et al. Altered sirtuin expression is associated with node-positive breast cancer. Br J Cancer 2006;95:1056–61.
[63] Aury-Landas J, Bougeard G, Castel H, Hernandez-Vargas H, Drouet A, Latouche JB, et al. Germline copy number variation of genes involved in chromatin remodelling in families suggestive of Li-Fraumeni syndrome with brain tumours. Eur J Human Genet 2013;21:1369–76.
[64] Laurent G, German NJ, Saha AK, de Boer VC, Davies M, Koves TR, et al. SIRT4 coordinates the balance between lipid synthesis and catabolism by repressing malonyl CoA decarboxylase. Mol Cell 2013;50:686–98.
[65] Laurent G, de Boer VC, Finley LW, Sweeney M, Lu H, Schug TT, et al. SIRT4 represses peroxisome proliferator-activated receptor alpha activity to suppress hepatic fat oxidation. Mol Cell Biol 2013;33:4552–61.
[66] Ho L, Titus AS, Banerjee KK, George S, Lin W, Deota S, et al. SIRT4 regulates ATP homeostasis and mediates a retrograde signaling via AMPK. Aging (Albany, NY) 2013;5:835–49.
[67] Mathias RA, Greco TM, Oberstein A, Budayeva HG, Chakrabarti R, Rowland EA, et al. Sirtuin 4 is a lipoamidase regulating pyruvate dehydrogenase complex activity. Cell 2014;159:1615–25.
[68] Blaveri E, Simko JP, Korkola JE, Brewer JL, Baehner F, Mehta K, et al. Bladder cancer outcome and subtype classification by gene expression. Clin Cancer Res 2005;11:4044–55.
[69] Garber ME, Troyanskaya OG, Schluens K, Petersen S, Thaesler Z, Pacyna-Gengelbach M, et al. Diversity of gene expression in adenocarcinoma of the lung. Proc Natl Acad Sci USA 2001;98:13784–9.
[70] Wang Q, Wen YG, Li DP, Xia J, Zhou CZ, Yan DW, et al. Upregulated INHBA expression is associated with poor survival in gastric cancer. Med Oncol 2012;29:77–83.
[71] Jeong SM, Lee A, Lee J, Haigis MC. SIRT4 protein suppresses tumor formation in genetic models of Myc-induced B cell lymphoma. J Biol Chem 2014;289:4135–44.
[72] Metallo CM, Gameiro PA, Bell EL, Mattaini KR, Yang J, Hiller K, et al. Reductive glutamine metabolism by IDH1 mediates lipogenesis under hypoxia. Nature 2012;481:380–4.
[73] Mullen AR, Wheaton WW, Jin ES, Chen PH, Sullivan LB, Cheng T, et al. Reductive carboxylation supports growth in tumour cells with defective mitochondria. Nature 2012;481:385–8.

Please cite this article in press as: Sebastián C, Mostoslavsky R. The role of mammalian sirtuins in cancer metabolism. Semin Cell Dev Biol (2015), http://dx.doi.org/10.1016/j.semcdb.2015.07.008

[74] Wise DR, DeBerardinis RJ, Mancuso A, Sayed N, Zhang XY, Pfeiffer HK, et al. Myc regulates a transcriptional program that stimulates mitochondrial glutaminolysis and leads to glutamine addiction. Proc Natl Acad Sci USA 2008;105:18782–7.
[75] Gao P, Tchernyshyov I, Chang TC, Lee YS, Kita K, Ochi T, et al. c-Myc suppression of miR-23a/b enhances mitochondrial glutaminase expression and glutamine metabolism. Nature 2009;458:762–5.
[76] Choo AY, Kim SG, Vander Heiden MG, Mahoney SJ, Vu H, Yoon SO, et al. Glucose addiction of TSC null cells is caused by failed mTORC1-dependent balancing of metabolic demand with supply. Mol Cell 2010;38:487–99.
[77] Wang L, Zhou H, Wang Y, Cui G, Di LJ. CtBP maintains cancer cell growth and metabolic homeostasis via regulating SIRT4. Cell Death Dis 2015;6:e1620.
[78] Frye RA. Phylogenetic classification of prokaryotic and eukaryotic Sir2-like proteins. Biochem Biophys Res Commun 2000;273:793–8.
[79] Park J, Chen Y, Tishkoff DX, Peng C, Tan M, Dai L, et al. SIRT5-mediated lysine desuccinylation impacts diverse metabolic pathways. Mol Cell 2013;50:919–30.
[80] Nakagawa T, Lomb DJ, Haigis MC, Guarente L. SIRT5 Deacetylates carbamoyl phosphate synthetase 1 and regulates the urea cycle. Cell 2009;137:560–70.
[81] Ogura M, Nakamura Y, Tanaka D, Zhuang X, Fujita Y, Obara A, et al. Overexpression of SIRT5 confirms its involvement in deacetylation and activation of carbamoyl phosphate synthetase 1. Biochem Biophys Res Commun 2010;393:73–8.
[82] Polletta L, Vernucci E, Carnevale I, Arcangeli T, Rotili D, Palmerio S, et al. SIRT5 regulation of ammonia-induced autophagy and mitophagy. Autophagy 2015;11:253–70.
[83] Lin ZF, Xu HB, Wang JY, Lin Q, Ruan Z, Liu FB, et al. SIRT5 desuccinylates and activates SOD1 to eliminate ROS. Biochem Biophys Res Commun 2013;441:191–5.
[84] Papa L, Hahn M, Marsh EL, Evans BS, Germain D. SOD2 to SOD1 switch in breast cancer. J Biol Chem 2014;289:5412–6.
[85] Somwar R, Erdjument-Bromage H, Larsson E, Shum D, Lockwood WW, Yang G, et al. Superoxide dismutase 1 (SOD1) is a target for a small molecule identified in a screen for inhibitors of the growth of lung adenocarcinoma cell lines. Proc Natl Acad Sci USA 2011;108:16375–80.
[86] Glasauer A, Sena LA, Diebold LP, Mazar AP, Chandel NS. Targeting SOD1 reduces experimental non-small-cell lung cancer. J Clin Invest 2014;124:117–28.
[87] Baysal BE, Ferrell RE, Willett-Brozick JE, Lawrence EC, Myssiorek D, Bosch A, et al. Mutations in SDHD, a mitochondrial complex II gene, in hereditary paraganglioma. Science 2000;287:848–51.
[88] N. Cancer Genome Atlas Research, Integrated genomic analyses of ovarian carcinoma, Nature 474 (2011) 609–615.
[89] Lu W, Zuo Y, Feng Y, Zhang M. SIRT5 facilitates cancer cell growth and drug resistance in non-small cell lung cancer. Tumour Biol 2014;35:10699–705.
[90] Michishita E, McCord RA, Berber E, Kioi M, Padilla-Nash H, Damian M, et al. SIRT6 is a histone H3 lysine 9 deacetylase that modulates telomeric chromatin. Nature 2008;452:492–6.
[91] Michishita E, McCord RA, Boxer LD, Barber MF, Hong T, Gozani O, et al. Cell cycle-dependent deacetylation of telomeric histone H3 lysine K56 by human SIRT6. Cell Cycle 2009;8:2664–6.
[92] Yang B, Zwaans BM, Eckersdorff M, Lombard DB. The sirtuin SIRT6 deacetylates H3 K56Ac in vivo to promote genomic stability. Cell Cycle 2009;8:2662–3.
[93] Gil R, Barth S, Kanfi Y, Cohen HY. SIRT6 exhibits nucleosome-dependent deacetylase activity. Nucleic Acids Res 2013;41:8537–45.
[94] Mao Z, Hine C, Tian X, Van Meter M, Au M, Vaidya A, et al. SIRT6 promotes DNA repair under stress by activating PARP1. Science 2011;332:1443–6.
[95] Van Meter M, Kashyap M, Rezazadeh S, Geneva AJ, Morello TD, Seluanov A, et al. SIRT6 represses LINE1 retrotransposons by ribosylating KAP1 but this repression fails with stress and age. Nat Commun 2014;5:5011.
[96] Mostoslavsky R, Chua KF, Lombard DB, Pang WW, Fischer MR, Gellon L, et al. Genomic instability and aging-like phenotype in the absence of mammalian SIRT6. Cell 2006;124:315–29.
[97] Kaidi A, Weinert BT, Choudhary C, Jackson SP. Human SIRT6 promotes DNA end resection through CtIP deacetylation. Science 2010;329:1348–53.
[98] McCord RA, Michishita E, Hong T, Berber E, Boxer LD, Kusumoto R, et al. SIRT6 stabilizes DNA-dependent protein kinase at chromatin for DNA double-strand break repair. Aging (Albany NY) 2009;1:109–21.
[99] Toiber D, Erdel F, Bouazoune K, Silberman DM, Zhong L, Mulligan P, et al. SIRT6 recruits SNF2H to DNA break sites, preventing genomic instability through chromatin remodeling. Mol Cell 2013;51:454–68.
[100] Kim HS, Xiao C, Wang RH, Lahusen T, Xu X, Vassilopoulos A, et al. Hepatic-specific disruption of SIRT6 in mice results in fatty liver formation due to enhanced glycolysis and triglyceride synthesis. Cell Metab 2010;12:224–36.
[101] Xiong X, Tao R, DePinho RA, Dong XC. Deletion of hepatic FoxO1/3/4 genes in mice significantly impacts on glucose metabolism through downregulation of gluconeogenesis and upregulation of glycolysis. PLoS ONE 2013;8:e74340.
[102] Zhong L, D'Urso A, Toiber D, Sebastian C, Henry RE, Vadysirisack DD, et al. The histone deacetylase Sirt6 regulates glucose homeostasis via Hif1alpha. Cell 2010;140:280–93.
[103] Kugel S, Mostoslavsky R. Chromatin and beyond: the multitasking roles for SIRT6. Trends Biochem Sci 2014;39:72–81.
[104] Dominy Jr JE, Lee Y, Jedrychowski MP, Chim H, Jurczak MJ, Camporez JP, et al. The deacetylase Sirt6 activates the acetyltransferase GCN5 and suppresses hepatic gluconeogenesis. Mol Cell 2012;48:900–13.
[105] Lyssiotis CA, Cantley LC. SIRT6 puts cancer metabolism in the driver's seat. Cell 2012;151:1155–6.
[106] Lai CC, Lin PM, Lin SF, Hsu CH, Lin HC, Hu ML, et al. Altered expression of SIRT gene family in head and neck squamous cell carcinoma. Tumour Biol 2013;34:1847–54.
[107] Marquardt JU, Fischer K, Baus K, Kashyap A, Ma S, Krupp M, et al. Sirtuin-6-dependent genetic and epigenetic alterations are associated with poor clinical outcome in hepatocellular carcinoma patients. Hepatology 2013;58:1054–64.
[108] Min L, Ji Y, Bakiri L, Qiu Z, Cen J, Chen X, et al. Liver cancer initiation is controlled by AP-1 through SIRT6-dependent inhibition of survivin. Nat Cell Biol 2012;14:1203–11.
[109] Lin Z, Yang H, Tan C, Li J, Liu Z, Quan Q, et al. USP10 antagonizes c-Myc transcriptional activation through SIRT6 stabilization to suppress tumor formation. Cell Rep 2013;5:1639–49.
[110] Kim EJ, Juhnn YS. Cyclic AMP signaling reduces sirtuin 6 expression in non-small cell lung cancer cells by promoting ubiquitin-proteasomal degradation via inhibition of the Raf-MEK-ERK (Raf/Mitogen-activated extracellular signal-regulated kinase/extracellular signal-regulated kinase) pathway. J Biol Chem 2015;290:9604–13.
[111] Choe M, Brusgard JL, Chumsri S, Bhandary L, Zhao XF, Lu S, et al. The RUNX2 transcription factor negatively regulates SIRT6 expression to alter glucose metabolism in breast cancer cells. J Cell Biochem 2015, http://dx.doi.org/10.1002/jcb.25171, accepted for publication.
[112] Wu M, Seto E, Zhang J. E2F1 enhances glycolysis through suppressing Sirt6 transcription in cancer cells. Oncotarget 2015;6(13):11252–63.
[113] Bauer I, Grozio A, Lasiglie D, Basile G, Sturla L, Magnone M, et al. The NAD^+-dependent histone deacetylase SIRT6 promotes cytokine production and migration in pancreatic cancer cells by regulating Ca^{2+} responses. J Biol Chem 2012;287:40924–37.
[114] Khongkow M, Olmos Y, Gong C, Gomes AR, Monteiro LJ, Yague E, et al. SIRT6 modulates paclitaxel and epirubicin resistance and survival in breast cancer. Carcinogenesis 2013;34:1476–86.
[115] Liu Y, Xie QR, Wang B, Shao J, Zhang T, Liu T, et al. Inhibition of SIRT6 in prostate cancer reduces cell viability and increases sensitivity to chemotherapeutics. Prot Cell 2013;4(9):702–10.
[116] Martinez-Pastor B, Mostoslavsky R. Sirtuins, metabolism, and cancer, Front. Pharmacol 2012;3:22.
[117] Dioum EM, Chen R, Alexander MS, Zhang Q, Hogg RT, Gerard RD, et al. Regulation of hypoxia-inducible factor 2alpha signaling by the stress-responsive deacetylase sirtuin 1. Science 2009;324:1289–93.
[118] Lim JH, Lee YM, Chun YS, Chen J, Kim JE, Park JW. Sirtuin 1 modulates cellular responses to hypoxia by deacetylating hypoxia-inducible factor 1alpha. Mol Cell 2010;38:864–78.
[119] Laemmle A, Lechleiter A, Roh V, Schwarz C, Portmann S, Furer C, et al. Inhibition of SIRT1 impairs the accumulation and transcriptional activity of HIF-1alpha protein under hypoxic conditions. PLoS ONE 2012;7:e33433.
[120] Liu G, Bi Y, Shen B, Yang H, Zhang Y, Wang X, et al. SIRT1 limits the function and fate of myeloid-derived suppressor cells in tumors by orchestrating HIF-1alpha-dependent glycolysis. Cancer Res 2014;74:727–37.
[121] Yoon H, Shin SH, Shin DH, Chun YS, Park JW. Differential roles of Sirt1 in HIF-1alpha and HIF-2alpha mediated hypoxic responses. Biochem Biophys Res Commun 2014;444:36–43.
[122] Mao B, Zhao G, Lv X, Chen HZ, Xue Z, Yang B, et al. Sirt1 deacetylates c-Myc and promotes c-Myc/Max association. Int J Biochem Cell Biol 2011;43:1573–81.
[123] Yuan J, Minter-Dykhouse K, Lou Z. A c-Myc-SIRT1 feedback loop regulates cell growth and transformation. J Cell Biol 2009;185:203–11.
[124] Menssen A, Hydbring P, Kapelle K, Vervoorts J, Diebold J, Luscher B, et al. The c-MYC oncoprotein, the NAMPT enzyme, the SIRT1-inhibitor DBC1, and the SIRT1 deacetylase form a positive feedback loop. Proc Natl Acad Sci USA 2012;109:E187–96.
[125] Marshall GM, Liu PY, Gherardi S, Scarlett CJ, Bedalov A, Xu N, et al. SIRT1 promotes N-Myc oncogenesis through a positive feedback loop involving the effects of MKP3 and ERK on N-Myc protein stability. PLoS Genet 2011;7:e1002135.
[126] Vettraino M, Manerba M, Govoni M, Di Stefano G. Galloflavin suppresses lactate dehydrogenase activity and causes MYC downregulation in Burkitt lymphoma cells through NAD/NADH-dependent inhibition of sirtuin-1. Anti-Cancer Drugs 2013;24:862–70.
[127] Lan F, Cacicedo JM, Ruderman N, Ido Y. SIRT1 modulation of the acetylation status, cytosolic localization, and activity of LKB1. Possible role in AMP-activated protein kinase activation. J Biol Chem 2008;283:27628–35.
[128] Hardie DG. AMP-activated protein kinase: a cellular energy sensor with a key role in metabolic disorders and in cancer. Biochem Soc Trans 2011;39:1–13.
[129] Sanders MJ, Grondin PO, Hegarty BD, Snowden MA, Carling D. Investigating the mechanism for AMP activation of the AMP-activated protein kinase cascade. Biochem J 2007;403:139–48.
[130] Faubert B, Boily G, Izreig S, Griss T, Samborska B, Dong Z, et al. AMPK is a negative regulator of the Warburg effect and suppresses tumor growth in vivo. Cell Metab 2013;17:113–24.

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[131] Kim HS, Vassilopoulos A, Wang RH, Lahusen T, Xiao Z, Xu X, et al. SIRT2 maintains genome integrity and suppresses tumorigenesis through regulating APC/C activity. Cancer Cell 2011;20:487–99.
[132] Serrano L, Martinez-Redondo P, Marazuela-Duque A, Vazquez BN, Dooley SJ, Voigt P, et al. The tumor suppressor SirT2 regulates cell cycle progression and genome stability by modulating the mitotic deposition of H4K20 methylation. Genes Dev 2013;27:639–53.
[133] Almeida A, Bolanos JP, Moncada S. E3 ubiquitin ligase APC/C-Cdh1 accounts for the Warburg effect by linking glycolysis to cell proliferation. Proc Natl Acad Sci USA 2010;107:738–41.
[134] Seo KS, Park JH, Heo JY, Jing K, Han J, Min KN, et al. SIRT2 regulates tumour hypoxia response by promoting HIF-1alpha hydroxylation. Oncogene 2015;34:1354–62.
[135] Hitosugi T, Zhou L, Elf S, Fan J, Kang HB, Seo JH, et al. Phosphoglycerate mutase 1 coordinates glycolysis and biosynthesis to promote tumor growth. Cancer Cell 2012;22:585–600.
[136] Xu Y, Li F, Lv L, Li T, Zhou X, Deng CX, et al. Oxidative stress activates SIRT2 to deacetylate and stimulate phosphoglycerate mutase. Cancer Res 2014;74:3630–42.
[137] Wang YP, Zhou LS, Zhao YZ, Wang SW, Chen LL, Liu LX, et al. Regulation of G6PD acetylation by SIRT2 and KAT9 modulates NADPH homeostasis and cell survival during oxidative stress. EMBO J 2014;33:1304–20.
[138] Liu PY, Xu N, Malyukova A, Scarlett CJ, Sun YT, Zhang XD, et al. The histone deacetylase SIRT2 stabilizes Myc oncoproteins. Cell Death Differ 2013;20:503–14.
[139] Barber MF, Michishita-Kioi E, Xi Y, Tasselli L, Kioi M, Moqtaderi Z, et al. SIRT7 links H3K18 deacetylation to maintenance of oncogenic transformation. Nature 2012;487:114–8.
[140] Kim W, Kim JE. SIRT7 an emerging sirtuin: deciphering newer roles. J Physiol Pharmacol 2013;64:531–4.
[141] Zhang S, Chen P, Huang Z, Hu X, Chen M, Hu S, et al. Sirt7 promotes gastric cancer growth and inhibits apoptosis by epigenetically inhibiting miR-34a. Sci Rep 2015;5:9787.
[142] Yu H, Ye W, Wu J, Meng X, Liu RY, Ying X, et al. Overexpression of sirt7 exhibits oncogenic property and serves as a prognostic factor in colorectal cancer. Clin Cancer Res 2014;20:3434–45.
[143] Hubbi ME, Hu H, Kshitiz, Gilkes DM, Semenza GL. Sirtuin-7 inhibits the activity of hypoxia-inducible factors. J Biol Chem 2013;288:20768–75.
[144] Shin J, He M, Liu Y, Paredes S, Villanova L, Brown K, et al. SIRT7 represses Myc activity to suppress ER stress and prevent fatty liver disease. Cell Rep 2013;5:654–65.
[145] A.D.N.J.D. Grey, Life span extension research and public debate: social considerations, Stud. Ethics Law Technol. 1 (2007), Article 5.
[146] Ben-Shmuel S, Rostoker R, Scheinman EJ, LeRoith D. Metabolic syndrome, type 2 diabetes, and cancer: epidemiology and potential mechanisms. In: Handbook of Experimental Pharmacology; 2015.
[147] Ligibel J. Lifestyle factors in cancer survivorship. J Clin Oncol 2012;30:3697–704.
[148] Trounce I, Byrne E, Marzuki S. Decline in skeletal muscle mitochondrial respiratory chain function: possible factor in ageing. Lancet 1989;1:637–9.
[149] Ross JM, Oberg J, Brene S, Coppotelli G, Terzioglu M, Pernold K, et al. High brain lactate is a hallmark of aging and caused by a shift in the lactate dehydrogenase A/B ratio. Proc Natl Acad Sci USA 2010;107:20087–92.
[150] El-Hassan A, Zubairu S, Hothersall JS, Greenbaum AL. Age-related changes in enzymes of rat brain. 1. Enzymes of glycolysis, the pentose phosphate pathway and lipogenesis. Enzyme 1981;26:107–12.
[151] Hagopian K, Ramsey JJ, Weindruch R. Influence of age and caloric restriction on liver glycolytic enzyme activities and metabolite concentrations in mice. Exp Gerontol 2003;38:253–66.
[152] Zhou Q, Lam PY, Han D, Cadenas E. Activation of c-Jun-N-terminal kinase and decline of mitochondrial pyruvate dehydrogenase activity during brain aging. FEBS Lett 2009;583:1132–40.
[153] Salminen A, Kaarniranta K. Glycolysis links p53 function with NF-kappaB signaling: impact on cancer and aging process. J Cell Physiol 2010;224:1–6.
[154] Yoshino J, Mills KF, Yoon MJ, Imai S. Nicotinamide mononucleotide, a key NAD(+) intermediate, treats the pathophysiology of diet- and age-induced diabetes in mice. Cell Metab 2011;14:528–36.
[155] Cohen HY, Miller C, Bitterman KJ, Wall NR, Hekking B, Kessler B, et al. Calorie restriction promotes mammalian cell survival by inducing the SIRT1 deacetylase. Science 2004;305:390–2.
[156] Canto C, Auwerx J. Caloric restriction. SIRT1 and longevity. Trends Endocrinol Metab 2009;20:325–31.
[157] Gomes AP, Price NL, Ling AJ, Moslehi JJ, Montgomery MK, Rajman L, et al. Declining NAD(+) induces a pseudohypoxic state disrupting nuclear-mitochondrial communication during aging. Cell 2013;155:1624–38.
[158] Lanza IR, Nair KS. Mitochondrial function as a determinant of life span. Pflugers Archiv: Eur J Physiol 2010;459:277–89.
[159] Lombard DB, Miller RA, Ageing:. Sorting out the sirtuins. Nature 2012;483:166–7.

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