

Metformin: The sweet link between tumor genetics and metabolism?

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Abstract

Introduction

Epidemiological studies have shown a correlation between insulin resistance, a marker of impaired glucose metabolism and metabolic syndrome, and malignancy development. Insulin resistance leads to a hyperinsulinemic state, which is thought to promote carcinogenesis via the direct effect of insulin on the insulin like growth factor 1 (IGF-1), an increase in IGF-1 synthesis, modulation of sex hormone availability, and finally through the resultant elevated glucose levels that promote inflammation and aid glycolysis in cancer cells. In addition to an increased incidence of cancer induction, insulin resistance has also been correlated with worse prognosis in cancer patients undergoing active treatment. Metformin is an oral agent that is widely used in the treatment of diabetes as it has been shown to sensitize cells to the effects of insulin. Several epidemiologic studies show a potential protective effect of metformin in diabetic patients. Preclinical studies have shown a direct inhibitory effect of metformin on cancer cell lines both in vitro and in vivo. This is thought to be mediated through multiple mechanisms, including effects on cellular metabolism via the AMP-activated protein kinase (AMPK) pathway, effects on cell cycle progression, and decreased cellular oxygen consumption. Though the data is conflicting, several retrospective

studies suggest an antitumor benefit of metformin in cancer patients undergoing active treatment. Several prospective studies examining the role of metformin as an adjunctive antineoplastic agent are currently ongoing. This review serves to explore the current role of metformin in cancer treatment and prevention of cancer, and to hypothesize that, in part, the effects of metformin may come from modulation of the metabolic environment of tumour cells.

Conclusion

Cancer cells and their inherent molecular pathways appear to be greatly influenced by the metabolic environment. Metformin modulates pathways their pathways directly, while indirectly affecting them through its effect on the metabolic environment. Clinical trials will tell us how effective it is at preventing and treating cancer. In the meantime, metformin remains a promising cancer treatment that links the roots of cancer as both a metabolic and genetic disease.

Introduction

The concept of cancer as a metabolic disease was born nearly a century ago through the work of Nobel Prize winner Otto Warburg, who proposed that a reliance of glucose and glycolysis for energy derivation was the hallmark of cancer cells.¹ Several decades later, the hypothesis of cancer as a genetic disease gained momentum, leaving cancer metabolism as an afterthought.

However, significant recent data has again revealed cancer to be at least partially a metabolic disease² that can be manipulated through dietary changes to affect glycolysis and tumour survival,³ paying homage to

Warburg's origin theory. As a result, the field of oncology has begun to view cancer and its treatment as both genetic and metabolic.⁴

The concept of insulin resistance, a hallmark of diabetes and metabolic syndrome, as a cancer risk factor is not a novel one. In fact, an increased risk of the development of cancer from insulin resistance has been observed in multiple epidemiological studies.

Cohort studies have found an increased incidence of several malignancies including those of the bladder, breast, colon, endometrium, liver and pancreas in patients with type II diabetes. A meta-analysis of 20 cohort and case control studies revealed a 20% increased risk of breast cancer for women with a pre-existing diagnosis of diabetes as compared to women without type II diabetes.⁵ In addition to an increased incidence of cancer, insulin resistance has been also correlated with worse outcomes in cancer patients.

Insulin resistance results in a plethora of physiologic and cellular events leading to metabolic disarray, obesity, hormonal disruption, elevated circulating glucose levels, and elevated serum insulin levels (Figure 1), all of which can lead to the induction and progression of cancer.^{6,7} As such, it would appear that insulin resistance is primarily a metabolic event, secondarily fuelling the molecular and genetic alterations that can lead to the progression of cancer. This review serves to explore the current role of metformin in cancer treatment and prevention.

Discussion

The authors have referenced some of their own studies in this review. These referenced studies have been conducted in accordance with the

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Declaration of Helsinki (1964) and the protocols of these studies have been approved by the relevant ethics committees related to the institution in which they were performed. All human subjects, in these referenced studies, gave informed consent to participate in these studies.

Insulin resistance and carcinogenesis

Several mechanisms have been proposed to explain the molecular methods through which hyperinsulinaemia secondary to insulin resistance may promote tumourigenesis (Figure 1). These can be divided into direct effects, resulting from stimulation of tyrosine kinase receptors with subsequent effects on cellular metabolism, and indirect effects via modulation of sex hormone availability.⁸

Insulin increases the hepatic synthesis of the cellular growth factor IGF-1, and decreases levels of IGF binding protein.⁹ This results in increased levels of bioavailable IGF-1, which can activate the IGF-1 receptor (IGF1R), leading to activation of pro-survival signalling pathways, ultimately resulting in cell growth and survival.¹⁰ Furthermore, insulin reduces sex hormone-binding globulin (SHBG) levels, which consequently leads to increased levels of unbound circulating bioavailable oestrogens,¹¹ a potential indirect mechanism for the increased risk of breast cancer and other oestrogen-dependent malignancies.⁷

Metformin and cancer – potential mechanisms of action

Metformin is an inexpensive and widely available drug with a well-established safety profile. It has been used for decades as first line therapy in the treatment of type II diabetes. Unlike sulfonylureas and other secretagogues, metformin does not affect pancreatic beta islet cells to increase insulin secretion, but rather increases the insulin sensitivity of target peripheral tissues.¹² Its action is mediated via activation of AMP-dependent protein kinase (AMPK), a

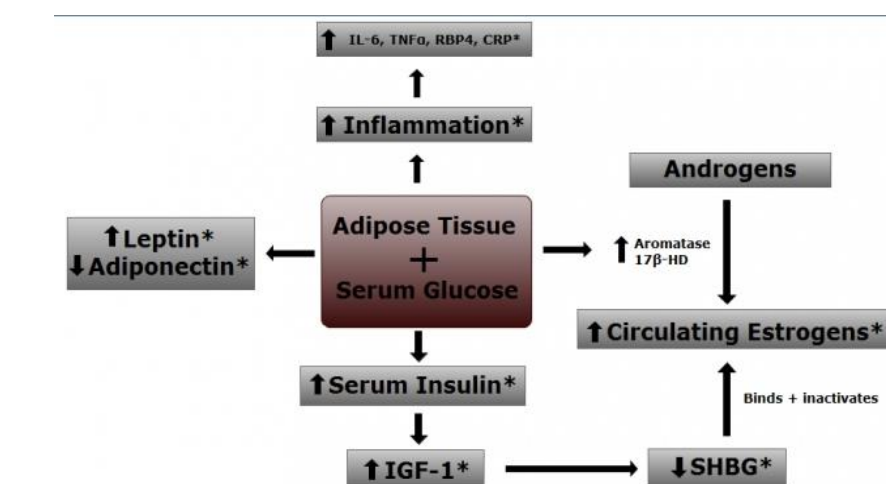


Figure 1: The metabolic effects of elevated glucose and excess adipose tissue. Both of these physiologic states may be mitigated through metformin usage. Legend: *represents a factor that is known to influence breast cancer recurrence. IGF-1: insulin-like growth factor 1 receptor, SHBG: sex hormone-binding globulin, TNF α : tumor necrosis factor alpha, IL-6: interleukin 6, RBP4: retinol-binding protein-4, CRP: C-reactive protein, 17 β -HD: 17 β -hydroxysteroid dehydrogenase. Image taken with permission from Champ et. al. (49).

metabolic regulator that is activated when the ratio of ATP to AMP becomes low, leads to mitochondrial biogenesis,¹³ increased glucose uptake, fatty acid oxidation and reduced gluconeogenesis in the liver.¹⁴

The resultant insulin sensitization may decrease levels of circulating insulin and counteract its tumorigenic activity, which could be exploited in cancer therapy. Additionally, this insulin sensitization will result in less circulating serum glucose levels, which can “feed” cancer cells, resulting in enhanced cell proliferation and the ability to overcome damage from chemotherapy and radiation therapy³.

Accordingly, there has been much interest in examining the indirect activity of metformin in cancer. Whereas increasing insulin sensitivity may indirectly prevent the tumorigenic effects of a hyperinsulinemic state, metformin also has direct effects that could be exploited against cancer growth. Metformin has a direct effect on cell metabolism and growth via activation of the AMPK pathway¹⁵ and subsequent downstream inhibition of mTOR.¹⁶

Metformin is also thought to lead to decreased oxygen consumption via the interaction with mitochondrial complex 1.¹⁷ Several in vitro studies support an anti-tumourigenic activity of metformin beyond its regulation of AMPK. A study examining the effects of metformin on a prostate cancer cell line showed that metformin led to the inhibition of cell proliferation. This effect was apparently not the result of increased apoptosis, as metformin decreased levels of cyclin D1 and cell cycle arrest.

Interestingly, this was observed even in the presence of AMPK inhibition, which is thought to be the major intracellular effector of metformin, revealing that there must be other pathways through which metformin leads to anti-proliferative effects in the cell.¹⁴ Other data in oestrogen receptor (ER) positive and human epidermal growth factor receptor 2 (Her2) positive breast cancer cell lines yielded similar results, with metformin-treated cells exhibiting a significant decrease in cyclin D1 and E2F1 with subsequent cell cycle arrest. In addition, in Her2 positive cell lines, treatment with metformin (with concentrations equivalent to the therapeutic range for diabetic patients) resulted in decreased Erb-B2 expression and decreased levels of activated Erb-B2, protein kinase B

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(Akt), and the mammalian target of rapamycin (mTOR), which have pro-survival functions.¹⁸ Other studies again point to the metabolic regulation of metformin, as exposure of breast cancer cells to metformin leads to increased AMPK activity, with subsequent decrease in mTOR activation.¹⁴

Other preclinical studies have focused on the effect of metformin on cancer stem cells, as metformin led to selective decrease in the stem cell population in a breast cancer cell line when it was combined with doxorubicin. The two agents acted synergistically in inducing cell death among the tumour colonies. A similar effect was seen when the two agents were given in combination in an in vivo xenograft mouse model. Tumour recovery from the xenografts following treatment showed absence of the stem cell cancer cell population in the combined treatment group, whereas stem cells were detected in the tumours from mice treated with doxorubicin only, indicating a preferential action of metformin on the stem cell cancer population.¹⁹ The same group showed similar synergistic effects of metformin when combined with other chemotherapeutic agents such as carboplatin and paclitaxel in lung and prostate cancer cell lines.²⁰

Not surprisingly, there is preclinical evidence for synergistic effects of metformin with radiation therapy (RT) as well. Over a century of data has revealed decreases in tumour induction and growth with the reduction of calories and dietary carbohydrates, and the subsequent decrease in circulating insulin and enhanced insulin sensitivity (Figure 2), the same pathways down regulated by metformin.²¹ A recent randomized trial revealed a significant up regulation of the AMPK pathway with five days of carbohydrate restriction.²² More recent data has revealed decreased tumour growth and modulation of the insulin and AMPK pathways through dietary restriction in combination with RT.²³

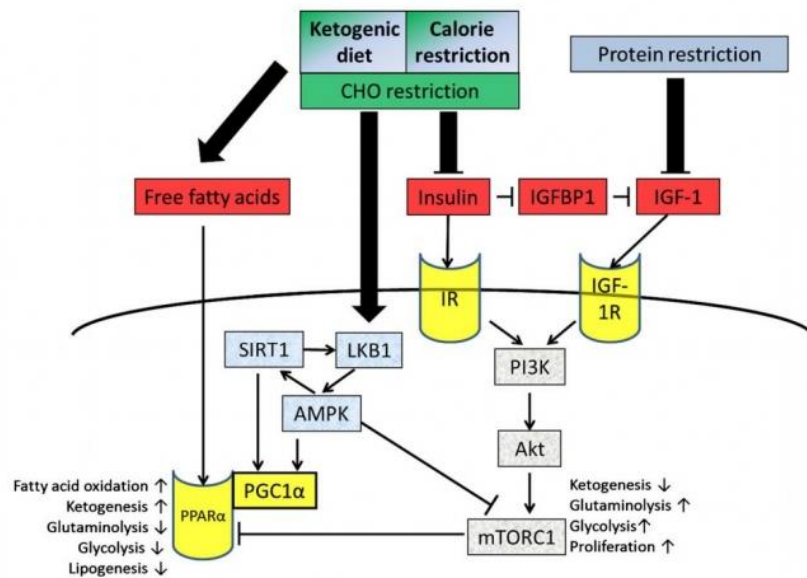


Figure 2: Dietary manipulation via calorie restriction, carbohydrate restriction, and a ketogenic diet target many of the same molecular pathways as those of metformin, upregulating the AMPK pathway. Image taken with permission from Klement and Champ (3).

A recent study examining the interactions of metformin with RT in hepatocellular carcinoma and normal hepatocyte cell lines showed decreased cell survival following irradiation in cells exposed to metformin prior to treatment.

Cells in the dual modality treatment showed increased levels of caspases, indicating a higher level of apoptotic cells post treatment. Greater amount and persistence following treatment of double stranded DNA breaks was also observed. In contrast, little interaction was seen between irradiation and metformin in normal hepatocytes.²⁴

Another study examining the interaction of metformin with irradiation in a breast cancer cell line showed decreased cell survival following irradiation in cells exposed to metformin prior to and after irradiation. In addition, metformin had a preferential cytotoxic effect on the stem cell population²⁵. Similar effects were seen in lung cancer cell lines, also revealing decreased cell survival following exposure prior to irradiation, with these findings confirmed in vivo in a mouse xenograft model.²⁶

A recent study analysing tumour hypoxia showed decreased oxygen consumption in several cancer cell lines in vitro following exposure to metformin. Moreover, in their in vivo model, this group demonstrated significant tumour reoxygenation in tumour grafts following treatment with metformin. In addition, in mice exposed to metformin prior to irradiation there was a significant tumour growth delay when compared to their non-metformin counterparts. They hypothesized that the increased oxygenation was secondary to inhibition of mitochondrial complex 1 by metformin, with resultant decreased oxygen consumption.²⁷

Metformin and cancer—clinical evidence

Several observational studies have noted a lower incidence of cancer and cancer-specific mortality in diabetic patients taking metformin, suggesting a protective effect. A meta-analysis of five observational studies revealed a 30% risk reduction in overall cancer incidence and mortality for diabetic patients on metformin therapy.²⁸ Similarly, a meta-analysis of 20 studies examined various malignancies and again showed a significant overall survival benefit (hazard ratio [HR]

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0.66) and cancer-specific survival benefit (HR 0.62) for diabetic patients on metformin.²⁹ Finally, data from the Women's Health Initiative, an observational study of nearly 70,000 postmenopausal women, found that metformin use in women with diabetes was associated with a lower incidence of invasive breast cancer, an observation that was noted for ER and progesterone receptor positive and Her2 negative cancers.³⁰

Other observational studies, however, have shown conflicting results regarding the benefit of metformin in cancer prevention.^{31,32,33,34,35,36,37,38} Yet, it stands to reason that increasing the insulin sensitivity of diabetic patients would provide a cancer-specific benefit by preventing the activation of the insulin pathway in cancer cells.

These observational studies of cancer incidence in patients on metformin have provided an impetus for further investigation. Data are now emerging which suggest metformin provides therapeutic benefits in patients with an established cancer diagnosis. Much of this data surrounds breast cancer, a disease that is associated with type II diabetes and obesity. For example, a 2009 retrospective study of over 2,500 patients analysed whether metformin would increase the efficacy of neoadjuvant chemotherapy in diabetic patients with breast cancer. Investigators found a threefold increase in pathologic complete response (pCR) in diabetic women on metformin therapy as compared to diabetic women not on metformin, as well as an 8% absolute increase in pCR when compared to non-diabetic women.³⁹

This antitumor benefit, however, was not reproduced in a series of over 1,400 women treated at MD Anderson Cancer Center, as metformin use during adjuvant chemotherapy did not significantly impact survival outcomes in diabetic patients with triple negative breast cancer.⁴⁰

There is a paucity of prospective data regarding metformin from

therapeutic, interventional breast cancer trials. Recent trials have examined biomarkers for evidence of anti-proliferative effects from pre-operative, oral metformin in women with breast cancer. Two phase II randomized trials assessing Ki67 levels compared metformin to a placebo prior to surgery in women with early stage breast cancer and have shown conflicting results.^{41,42}

In another randomized trial from Italy assessing four weeks of preoperative metformin, investigators compared apoptosis levels in core biopsies and surgical specimens but found no significant modulation of apoptosis by metformin, but response may have correlated with the degree of insulin resistance.⁴³ In view of the promising data from retrospective cohort studies, meta-analyses, observational studies, and limited early prospective investigations, metformin is currently being evaluated in a randomized trial as an adjuvant therapy for women with breast cancer.⁴⁴

Similar to breast cancer, obesity and insulin resistance strongly impact the pathogenesis of endometrial cancer, and hindering hyperinsulinemia with metformin may be useful as a treatment. Recently, two large patient series have illustrated the potential therapeutic benefits of metformin in endometrial cancer. Nevadunsky and colleagues evaluated the survival times of nearly one thousand patients with endometrial cancer, and found that the overall survival of diabetic patients who received metformin exceeded that of both non-diabetics and diabetics who did not receive metformin therapy.⁴⁵

Interestingly, this proposed trend was limited to women with type II non-endometrioid cancers which represented only a small patient subgroup. In a second large series, a pooled analysis of diabetic endometrial cancer patients on metformin had significantly improved recurrence-free and overall survival when compared to diabetics on other medications. However, time to cancer

recurrence was not improved, suggesting a benefit for all-cause mortality rather than cancer specific outcomes.⁴⁶

Data from an analysis of nearly 4,000 diabetic men with prostate cancer in a Canadian cancer registry revealed that men receiving metformin therapy for a median of 8.9 months after their cancer diagnosis had improved prostate cancer-specific survival and all-cause mortality.³⁷ These findings have stimulated the development of at least 10 clinical trials of metformin as a therapeutic strategy for prostate cancer including the Metformin Active Surveillance Trail (MAST) in Canada, which will randomize men with low risk prostate cancer seeking active surveillance to metformin or placebo control to determine whether time to progression can be delayed by metformin use.

Retrospective data in diabetic patients with pancreatic cancer revealed an increase in overall survival for those with non-metastatic disease receiving metformin.⁴⁷

Additionally, a recent retrospective study of diabetic patients with oesophageal adenocarcinoma showed that those patients on metformin had higher pCR (34.5%) and enhanced radiographic response rates as compared to patients with diabetes not on metformin following neoadjuvant concurrent chemo-radiation.

Interestingly, their response rates were even greater than those without diabetes. There was also an increase in locoregional control, but no difference in overall or progression free survival.⁴⁸

Current Issues

The available data on the benefits of metformin has many important limitations, including the inclusion of small patient subgroups with diabetes, the inconsistent definition of metformin users, the lack of data surrounding length of metformin use, the inherent flaws associated with retrospective analysis, and the heterogeneity of patients on metformin and their coexisting metabolic risk factors.

Additional research is needed to clarify whether the observed association between metformin use and cancer outcomes is the result of shared prognostic variables or whether the relationship is causal. The current data are only hypothesis generating and highlight the need for the nearly 100 prospective trials which are now being developed to investigate the antitumor effects of metformin in a wide array of malignancies.

Metformin will be investigated in both the setting of primary and secondary prevention. While it is unknown at this time which population, if any, will realize a benefit, the current evidence suggests a potential future role for metformin in the treatment and prevention of malignancies.

Conclusion

While it remains unclear if cancer is primarily a genetic or metabolic disease, metformin may reveal that it is both. Preclinical data has exposed a potential usage for metformin for cancer prevention or to enhance current cancer therapies like chemotherapy and radiation therapy by upregulation of the AMPK pathway, with down regulation of the insulin, IGF-1, and mTOR pathway. Metformin may simply be reflecting a century of data on the effect of dietary manipulation on cancer survival by affecting the same pathways.

Consequently, current data has revealed the detrimental effects of obesity, insulin resistance, and chronically elevated serum glucose levels on cancer occurrence and poorer outcomes after treatment. As rates of metabolic dysfunction and obesity continue to rise, the oncologist is forced to ask whether metformin is treating the cancer cells, or repairing the metabolic dysfunction with which many patients are currently experiencing.

Future randomized trials will assess metformin's action in many patient subgroups to fully manipulate the

metabolic and cellular survival mechanisms of cancer and answer many of these questions. Until then, metformin remains a promising cancer treatment that links the roots of cancer as both a metabolic and genetic disease.

References

1. Warburg O, Wind F, Negelein E. The Metabolism of Tumors in the Body. *J. Gen. Physiol.* 1927;8:519-530. Available at: <http://www.ncbi.nlm.nih.gov/pubmed/19872213>.
2. Seyfried TN. *Cancer as a Metabolic Disease: On the Origin, Management, and Prevention of Cancer.* John Wiley & Sons; 2012:438.
3. Klement RJ, Champ CE. Calories, carbohydrates, and cancer therapy with radiation: exploiting the five R's through dietary manipulation. *Cancer Metastasis Rev.* 2014.
4. Champ CE, Palmer JD, Volek JS, et al. Targeting metabolism with a ketogenic diet during the treatment of glioblastoma multiforme. *J. Neurooncol.* 2014;117(1):125-31.
5. Larsson SC, Mantzoros CS, Wolk A. Diabetes mellitus and risk of breast cancer: a meta-analysis. *Int. J. Cancer.* 2007;121(4):856-62.
6. Calle EE, Kaaks R. Overweight, obesity and cancer: epidemiological evidence and proposed mechanisms. *Nat Rev Cancer.* 2004;4(8):579-591.
7. Champ CE, Volek JS, Siglin J, Jin L, Simone NL. Weight Gain, Metabolic Syndrome, and Breast Cancer Recurrence: Are Dietary Recommendations Supported by the Data? *Int. J. Breast Cancer.* 2012;2012:9.
8. Arcidiacono B, Iiritano S, Nocera A, et al. Insulin resistance and cancer risk: an overview of the pathogenetic mechanisms. *Exp. Diabetes Res.* 2012;2012:789174.
9. Scharf J, Ramadori G, Braulke T, Hartmann H. Synthesis of insulinlike growth factor binding proteins and of the acid-labile subunit in primary cultures of rat hepatocytes, of Kupffer cells, and in cocultures: regulation by insulin, insulinlike growth factor, and growth hormone. *Hepatology.* 1996; 23(4):818-27.
10. Pollak M. Insulin and insulin-like growth factor signalling in neoplasia. *Nat. Rev. Cancer.* 2008;8(12):915-28.
11. Crave JC, Lejeune H, Brébant C, Baret C, Pugeat M. Differential effects of insulin and insulin-like growth factor I on the production of plasma steroid-binding globulins by human hepatoblastoma-derived (Hep G2) cells. 2013. Available at: <http://press.endocrine.org/doi/abs/10.1210/jcem.80.4.7536204>. Accessed April 2, 2014.
12. Goodman and Gilman's *The Pharmacological Basis of Therapeutics*, Twelfth Edition - Laurence Brunton, Bruce Chabner, Bjorn Knollman. Available at: <http://www.mcgraw-hill.com.au/html/9780071624428.html>. Accessed April 2, 2014.
13. Zong H, Ren JM, Young LH, et al. AMP kinase is required for mitochondrial biogenesis in skeletal muscle in response to chronic energy deprivation. *Proc. Natl. Acad. Sci. U. S. A.* 2002;99(25):15983-7.
14. Zakikhani M, Dowling R, Fantus IG, Sonenberg N, Pollak M. Metformin is an AMP kinase-dependent growth inhibitor for breast cancer cells. *Cancer Res.* 2006;66(21):10269-73.
15. Owen MR, Doran E, Halestrap AP. Evidence that metformin exerts its antidiabetic effects through inhibition of complex 1 of the mitochondrial respiratory chain. *Biochem. J.* 2000;348 Pt 3:607-14. Available at: <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=1221104&tool=pmcentrez&rendertype=abstract>. Accessed April 2, 2014.
16. Ben Sahra I, Laurent K, Loubat A, et al. The antidiabetic drug metformin exerts an antitumoral effect in vitro and in vivo through a decrease of cyclin D1 level. *Oncogene.* 2008; 27(25):3576-86.
17. Dowling RJO, Zakikhani M, Fantus IG, Pollak M, Sonenberg N. Metformin inhibits mammalian target of rapamycin-dependent translation initiation in breast cancer cells. *Cancer Res.* 2007;67(22):10804-12.
18. Alimova IN, Liu B, Fan Z, et al. Metformin inhibits breast cancer cell growth, colony formation and induces cell cycle arrest in vitro. *Cell Cycle.*

- 2009;8(6):909-15. Available at: <http://www.ncbi.nlm.nih.gov/pubmed/19221498>. Accessed April 2, 2014.
19. Hirsch HA, Iliopoulos D, Tsihchlis PN, Struhl K. Metformin selectively targets cancer stem cells, and acts together with chemotherapy to block tumor growth and prolong remission. *Cancer Res.* 2009;69(19):7507-11.
20. Iliopoulos D, Hirsch HA, Struhl K. Metformin decreases the dose of chemotherapy for prolonging tumor remission in mouse xenografts involving multiple cancer cell types. *Cancer Res.* 2011;71(9):3196-201.
21. Champ CE, Baserga R, Mishra M V, et al. Nutrient Restriction and Radiation Therapy for Cancer Treatment: When Less Is More. *Oncologist.* 2013;18(1):97-103.
22. Draznin B, Wang C, Adochio R, Leitner JW, Cornier MA. Effect of Dietary Macronutrient Composition on AMPK and SIRT1 Expression and Activity in Human Skeletal Muscle. *Horm Metab Res.* 2012;44(09):650-655.
23. Saleh AD, Simone BA, Palazzo J, et al. Caloric restriction augments radiation efficacy in breast cancer. *Cell cycle Georg. Tex.* 2013;12(12):1955-1963.
24. Kim EH, Kim M-S, Cho C-K, Jung W-G, Jeong YK, Jeong J-H. Low and high linear energy transfer radiation sensitization of HCC cells by metformin. *J. Radiat. Res.* 2013.
25. Song CW, Lee H, Dings RPM, et al. Metformin kills and radiosensitizes cancer cells and preferentially kills cancer stem cells. *Sci Rep.* 2012;2:362.
26. Storzuk Y, Hopmans SN, Sanli T, et al. Metformin inhibits growth and enhances radiation response of non-small cell lung cancer (NSCLC) through ATM and AMPK. *Br. J. Cancer.* 2013;108(10):2021-32.
27. Zannella VE, Dal Pra A, Muaddi H, et al. Reprogramming metabolism with metformin improves tumor oxygenation and radiotherapy response. *Clin. Cancer Res.* 2013;19(24):6741-50.
28. Decensi A, Puntoni M, Goodwin P, et al. Metformin and cancer risk in diabetic patients: a systematic review and meta-analysis. *Cancer Prev. Res. (Phila).* 2010;3(11):1451-61.
29. Yin M, Zhou J, Gorak EJ, Quddus F. Metformin is associated with survival benefit in cancer patients with concurrent type 2 diabetes: a systematic review and meta-analysis. *Oncologist.* 2013;18(12):1248-55.
30. Chlebowski RT, McTiernan A, Wactawski-Wende J, et al. Diabetes, Metformin, and Breast Cancer in Postmenopausal Women. *J. Clin. Oncol.* 2012;30(23):2844-52.
31. Bodmer M, Becker C, Meier C, Jick SS, Meier CR. Use of antidiabetic agents and the risk of pancreatic cancer: a case-control analysis. *Am. J. Gastroenterol.* 2012;107(4):620-6.
32. Bodmer M, Becker C, Meier C, Jick SS, Meier CR. Use of metformin and the risk of ovarian cancer: a case-control analysis. *Gynecol. Oncol.* 2011;123(2):200-4.
33. Becker C, Meier CR, Jick SS, Bodmer M. Case-control analysis on metformin and cancer of the esophagus. *Cancer Causes Control.* 2013;24(10):1763-70.
34. Becker C, Jick SS, Meier CR, Bodmer M. Metformin and the risk of endometrial cancer: a case-control analysis. *Gynecol. Oncol.* 2013;129(3):565-9.
35. Col NF, Ochs L, Springmann V, Aragaki AK, Chlebowski RT. Metformin and breast cancer risk: a meta-analysis and critical literature review. *Breast Cancer Res. Treat.* 2012;135(3):639-46.
36. Smiechowski BB, Azoulay L, Yin H, Pollak MN, Suissa S. The use of metformin and the incidence of lung cancer in patients with type 2 diabetes. *Diabetes Care.* 2013;36(1):124-9.
37. Margel D, Urbach D, Lipscombe LL, et al. Association between metformin use and risk of prostate cancer and its grade. *J. Natl. Cancer Inst.* 2013;105(15):1123-31.
38. Noto H, Goto A, Tsujimoto T, Noda M. Cancer risk in diabetic patients treated with metformin: a systematic review and meta-analysis. *PLoS One.* 2012;7(3):e33411.
39. Jiralerspong S, Palla SL, Giordano SH, et al. Metformin and pathologic complete responses to neoadjuvant chemotherapy in diabetic patients with breast cancer. *J. Clin. Oncol.* 2009;27(20):3297-302.
40. Bayraktar S, Hernandez-Aya LF, Lei X, et al. Effect of metformin on survival outcomes in diabetic patients with triple receptor-negative breast cancer. *Cancer.* 2012;118(5):1202-11.
41. Bonanni B, Puntoni M, Cazzaniga M, et al. Dual effect of metformin on breast cancer proliferation in a randomized presurgical trial. *J. Clin. Oncol.* 2012;30(21):2593-600.
42. Hadad S, Iwamoto T, Jordan L, et al. Evidence for biological effects of metformin in operable breast cancer: a pre-operative, window-of-opportunity, randomized trial. *Breast Cancer Res. Treat.* 2011;128(3):783-94.
43. Cazzaniga M, DeCensi A, Pruneri G, et al. The effect of metformin on apoptosis in a breast cancer presurgical trial. *Br. J. Cancer.* 2013;109(11):2792-7.
44. Goodwin PJ, Stambolic V, Lemieux J, et al. Evaluation of metformin in early breast cancer: a modification of the traditional paradigm for clinical testing of anti-cancer agents. *Breast Cancer Res. Treat.* 2010;126(1):215-220.
45. Nevadunsky NS, Van Arsdale A, Strickler HD, et al. Metformin use and endometrial cancer survival. *Gynecol. Oncol.* 2014;132(1):236-40.
46. Ko EM, Walter P, Jackson A, et al. Metformin is associated with improved survival in endometrial cancer. *Gynecol. Oncol.* 2014;132(2):438-42.
47. Sadeghi N, Abbruzzese JL, Yeung SJ, Hassan M, Li D. Effect of metformin on survival of diabetic patients with pancreatic adenocarcinoma. *ASCO Meet. Abstr.* 2011;29(15_suppl):4063. Available at: http://meeting.ascopubs.org/cgi/content/abstract/29/15_suppl/4063.
48. Skinner HD, McCurdy MR, Echeverria AE, et al. Metformin use and improved response to therapy in esophageal adenocarcinoma. *Acta Oncol.* 2013;52(5):1002-9.