ADDIS ABABA UNIVERSITY
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TITLE: **METFORMIN ANTICANCER ACTIVITY**

A REVIEW PAPER AND A METAANLYSIS SUBMITTED TO THE DEPARTMENT OF PHARMACOLOGY, SCHOOL OF MEDICINE, COLLEGE OF HEALTH SCIENCE, GRADUATE STUDY, ADDIS ABABA UNIVERSITY, IN PARTIAL FULFILMENT OF THE REQUIREMNETS FOR THE DEGREE OF MASTERS OF SCIENCE IN PHARMACOLOGY

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Acronyms

AMP…… Adenosine mono phosphate
AMPK……... Adenosine monophosphate kinase
DM…… Diabetes mellitus
DPP-4…… Dipeptidyl peptidase-4
ERK…….. Extracellular signal-regulated kinase
IGF-1…… Insulin like growth factor
IGFBP-1……Insulin like growth factor binding protein-1
mTOR……... Mammalian target of rapamycin
PI3K…….. Phosphoinositide3-kinase
T2DM……Type 2 diabetes mellitus
WHO……. World health organization

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Summary

Background: Accumulating evidence suggests that patients with type 2 diabetes mellitus (T2DM) and hyperinsulinemia are at increased risk for developing malignancies. It remains to be fully elucidated whether use of metformin, an insulin sensitizer, and/or sulfonylureas (SUs), insulin secretagogues, affect cancer incidence in subjects with T2DM.

Objective: A meta-analysis was performed to compare the risk of cancer incidence associated with monotherapy with SUs versus monotherapy with metformin in T2DM patients.

Material and Methods: Search was performed throughout MedLINE/PubMed, and Cochrane Central Register of Controlled Trials (CENTRAL) and ClinicalTrials.gov up to April 30, 2012. Fixed- and random-effect models basing on heterogeneity were fitted to estimate the summary relative risk (RR). Between studies heterogeneity was tested using X^2 statistics and measured with the I^2 statistic. Publication bias was evaluated using funnel plot and Egger's regression asymmetry test.

Results: A total of 10 studies, all of them were cohort studies, included in the meta-analysis. Obvious heterogeneity was noted, and Pooling of the raw data of those 10 cohort studies suggested that T2DM patients using SUs compared with metformin use had a significantly higher risk of overall cancer (RR= 1.59, 95% CI 1.38-1.83, I^2=94.88%, p< 0.00001), using the random-effects model. But, after pooling of the adjusted estimates, the use of SUs was not associated with a higher cancer risk than metformin treatment (RR=1.12, 95% CI 1.05–1.19, I^2=51.20%, p< 0.000145), using the random-effects model.

Conclusions and Recommendations: This analysis clearly shows that monotherapy with SUs can increase cancer risk compared to metformin monotherapy in T2DM patients. The evidence is not yet adequate to establish the effect of SUs, relative to metformin on cancer. Future investigators should consider more rigorous study design and data analysis, and use data with acceptable rigor. These findings need to be confirmed in large-scale RCTs before they are translated into clinical practice.

1. INTRODUCTION

1.1. Background

Type 2 diabetes mellitus (T2DM) is a progressive metabolic disorder that accounts for more than 90% of all cases of DM [1]. It is a chronic metabolic disorder characterized mainly by the presence of insulin resistance, diminished pancreatic beta cell production and persistently elevated blood glucose level [2]. The incidence and prevalence of T2DM has been increasing worldwide and currently considered as major health problem. Globally, an estimated 422 million adults were living with diabetes in 2014, compared to 108 million in 1980 [3].

A central aspect of type 2 DM is the development of chronic complications associated with high morbidity and mortality. Microvascular complications such as retinopathy, nephropathy and neuropathy are major causes of new cases of blindness and renal insufficiency. Moreover, macrovascular complications like peripheral arterial disease, coronary arterial disease and stroke are major causes of morbidity and mortality [4]. Besides, there is a growing evidence base to support a connection between diabetes, predominantly T2DM, and certain types of cancer [5]. In fact, meta-analyses have revealed T2DM to be an independent risk factor for the development of several different types of cancer. As yet, few studies have explored a relationship between type 1 diabetes mellitus and cancer [6].

1.2. Type 2 diabetes mellitus and cancer

Cancer is defined as not one but an array of diseases with more than 200 different types depending on the cell and tissue of origin. According to the most recent data released by the World Health Organization (WHO) in 2012, more than 14 million of new cancer cases were diagnosed, and 8.2 million cancer deaths and 32.4 million people living with cancer (within 5 years of diagnosis) were registered worldwide [7]. Although significant progress has been made in understanding the genetic and epigenetic factors that promotes tumor growth and metastasis, the concomitant progress in developing new cancer drugs is much slower.

The progression from a normal cell to a cancerous cell is a multistep process that reflects the accumulation of genetic and epigenetic alterations within a cell's genome. In general, high-frequency

mutations in cancer are mainly found in proto-oncogenes and tumor suppressor genes. Gain-of-function (GOF) mutations in proto-oncogenes result in hyperactive forms, termed oncogenes that can promote cell transformation. Conversely, loss-of-function (LOF) mutations in tumor suppressor genes, expression of which usually restricts cell transformation, result in a hypoactive or inactive state of the respective gene. Complex alterations occur during the process of transformation and some cancer cells become dependent on the continued activation of a single gene/pathway. This dependence is termed ‘oncogene addiction’ [8]. Moreover, Hanahan and Weinberg [9] reported six essential hallmarks that represent the essential alterations in cancer cell physiology: self-sufficiency in growth signals, tissue invasion and metastasis, evasion of apoptosis, sustained angiogenesis, limitless replicative potential, and insensitivity to anti-growth signals were described.

A number of large-scale epidemiologic studies and meta-analyses have shown a consistent increase in site-specific cancer incidence among patients with T2DM. This includes a two- to three-fold increase in the incidence rate of pancreatic cancer, a two-fold increased risk for hepatobiliary cancers, a 20% increased risk for breast cancer, a two-fold increased risk for endometrial cancer, and a 50% increased incidence of colorectal cancer [10]. It has also been demonstrated that patients with T2DM have excess mortality for a number of cancers, including a 30% to 40% increase with pancreatic, a 2.5-fold increase with liver, a 30% increase with endometrial, a 15% to 30% increase with breast cancer, and a 20% to 50% increase with colorectal cancer [11].

Mechanisms involved in the association between T2DM and cancer are not completely understood, but a number of factors have been proposed to contribute to the increased risk of cancer development and mortality in the setting of T2DM. These include hyperglycemia, insulin resistance, hyperinsulinemia, increased insulin-like growth factor-1 (IGF-1) levels, dyslipidemia, inflammatory cytokines [12, 13]. Accordingly, numerous studies have reported an increased incidence of breast, endometrial, and colorectal cancers that are most significant within the first months after T2DM diagnosis and even in the prediabetes phase [5]. These results suggest that hyperinsulinemia, rather than hyperglycemia, in diabetes is associated with an increased risk of cancer [11].

In general, hyperinsulinaemia may arise as a result of exogenous insulin administration in T1DM and endogenous insulin release at early stages of T2DM to compensate for peripheral insulin resistance that develops after frequent exposure to high circulating levels of glucose and lipids in

obesity. Insulin binding to its own tyrosine kinase receptor (IR) activates the PI3K/Akt/mTOR and MAPK/ERK pathways to induce metabolic, anti-apoptotic and proliferative changes. Insulin also binds (with lower affinity) the highly related receptor IGF-1R that signals towards proliferation through MAPK/ERK. Insulin signaling differs in healthy and diabetic patients [12, 13], with insulin resistance being characterized by an altered metabolic branch that leaves mitogenic signaling unaffected [14]. Thus, excess insulin binding to IRs and IGF-1R in diabetes may lead to proliferative and anti-apoptotic effects mediated by PI3K-Akt/mTOR and Ras-MAPK signaling pathways that are frequently activated in human cancers [15]. Cancer cells also frequently overexpress IGF-1R and the uncommon form of insulin receptor IR-A that signal predominantly via MAPK to promote proliferation [14].

1.3. Anti-diabetic Treatment and Cancer

Anti-diabetic drugs, including oral antidiabetic agents and exogenous insulin, have been widely used for T2DM patients for decades. Oral antidiabetic agents are intended to normalize blood sugar levels through three main mechanisms of action: increasing the amount of insulin secreted by the pancreas (insulin secretagogues), increasing the sensitivity of target organs to insulin (insulin sensitizers), and decreasing the rate at which glucose is absorbed from the gastrointestinal tract (alpha-glucosidase inhibitors). Some new anti-diabetic drug classes, glucagon-like peptide-1 (GLP-1) analogs and dipeptidyl peptidase-4 (DPP-4) inhibitors, may increase incretin levels, but the experience with these new compounds is still quite limited [16, 17].

Most T2DM patients have long-term exposure to multiple anti-diabetic treatments. If hyper-insulinemia plays a role in increasing cancer risk in diabetic patients, it is reasonable to expect that treatments that increase endogenous (sulfonylurea and meglitinides) or exogenous insulin (insulin replacement) may be risk factors for cancer development, and in contrast, those treatments that decrease endogenous insulin (metformin) may not be risk factors or even play protective roles in cancer occurrence among diabetics. Nevertheless, exposure to antidiabetic drugs (type, time and doses) should be among the most important factors included in studies on diabetes-cancer associations, and studies that aim to elucidate the mechanisms that may connect each antidiabetic drug with cancers need strong support. The most frequently prescribed oral glucose lowering drugs for treatment of T2DM are metformin and sulfonylurea derivatives [18].

The biguanide metformin, an inexpensive and well tolerated oral antidiabetic agent, has been widely used for more than 30 years. Acting as an insulin sensitizer, it reduces serum insulin levels by promoting utilization of insulin in the liver and in muscle. In recent years metformin has been recommended as the first line drug for T2DM, especially in those who are overweight [16]. Whereas, sulfonylureas are insulin secretagogues, which stimulate endogenous insulin secretion through direct action on pancreatic beta cells [17]. Sulfonlyurea used to be the first line drug for T2DM. Starting in the early 2000s, evidence has shown that metformin provides better glucose control, and low incidence of hypoglycemia. Since then, sulfonylureas have only been used as first-line therapy in patients of normal weight, or in those who cannot tolerate metformin [16]. On the other hand, the antidiabetic drugs metformin and sulfonylurea and their association with risk and outcome of cancer related with T2DM have been extensively studied.

Evans *et al*. [19] were the first to report a potential association of metformin use with reduced cancer incidence. The study result suggested that the use of metformin was associated with a 23% decreased risk of any cancer. Since then, a large number of observational studies have been published with several substantiating a possible decreased incidence of cancer with this drug. For instance, in a study done by Bowker *et al*. [20] that included 10, 309 diabetic patients, compared the incidence of cancer during treatment, i.e., insulin, metformin, or sulfonylureas, for a period of 5 years. They found that patients treated with metformin had a significantly lower rate of cancer-related mortality compared with patients exposed to sulfonylureas or insulin. Indeed, the cancer mortality rate in percent (per 1,000 person-years of follow-up) was 6.3% and 9.7% for metformin and sulfonylurea cohorts, respectively, and insulin users had higher incidence of cancer-related mortality than patients not receiving insulin.

Similarly, Currie *et al.* [21] published a retrospective cohort study in the UK, which included more than 100,000 patients from primary care practices, of which 8,392 had type II diabetes. All patients with or without diabetes who developed a tumor were followed for up to 19 years. Diabetes was stratified by treatment regimen and Cox proportional hazard models were used to compare all case mortality from all cancers and from specific cancers. They found that cancer mortality increased in patients with diabetes compared to non-diabetic patients (hazard ratio [HR] 1.09, 95% CI 1.06–1.13). When analyzed according to type of cancer, mortality was increased in breast and prostate cancer but

was decreased in lung cancer. However diabetic patients had a lower mortality rate than non-diabetic patients if they were treated with metformin (HR 0.85, 95% CI 0.78–0.93).

Additionally, Ruiter *et al.* [22] have done a large cohort study which included over 2.5 million diabetic patients who were identified via pharmacy dispensing records. They found that patients using metformin had a lower incidence of cancer compared to those who used sulfonylurea (HR 0.90, 95% CI 0.88–0.91). Besides, Chlebowski *et al*. [23] assessed the association between diabetes, metformin use, and breast cancer among 68,019 postmenopausal women participating in Women's Health Initiative clinical trials. As it was reported, compared with women without diabetes, in diabetic woman the incidence of breast cancer was related to diabetes therapy. Diabetic women not treated with metformin had a slightly higher incidence of breast cancer. The association was observed for cancers positive for estrogen and progesterone receptor as well as those negative for HER2. Moreover, recently Ki *et al.* [24] performed a retrospective cohort study, based on a nationwide population database, which included a total 4,503 patients. The study aimed at investigating the impact of metformin on the survival of patients with diabetes mellitus and non-metastatic rectal cancer who underwent curative surgery. From 4,503 patients, who were prescribed oral anti-diabetic agents, 3,694 patients received metformin for at least 90 days. Unadjusted analyses showed a significantly higher overall survival (OS) [HR, 0.596; 95% CI, 0.506 to 0.702] and rectal cancer-specific survival (HR, 0.621; 95% CI, 0.507 to 0.760) in the metformin group than in the non-metformin group. Meanwhile, the adjusted OS (HR, 0.631; 95% CI, 0.527 to 0.755) and cancer-specific survival (HR, 0.598; 95% CI, 0.479 to 0.746) in the group with a medication possession ratio of 80% or greater was significantly higher than in the group with a medication possession ratio of less than 80%.

Conversely, the positive correlation between metformin use and incidence of various type cancers was not universally noted by all investigators. Mamtani *et al*. [25] analyzed data from 87,600 patients with T2DM. They assessed the incidence of bladder cancer in new users of metformin and sulfonylurea and they reported no association between metformin use and this type cancer. Additionally, there are important differences in the characteristics of patients treated with metformin compared with other antidiabetic agents that may be responsible for the observed differences in cancer incidence. For instance, in the United Kingdom metformin users had a higher body mass

index, a younger age, a lower systolic BP, a lower prevalence of cardiovascular disease, and a higher proportion of aspirin use as compared with sulfonylurea users at the initiation of therapy [26].

To end with, the results from epidemiologic studies have been inconsistent, however, and the reasons underlying this heterogeneity, including differences between cancer sites, study populations, and design, need to be further investigated. In light of the increasing prevalence of patients under treatment with oral anti-diabetic drugs, efforts aimed at elucidating the association between use of these drugs and cancer risk have major implications for public health. With these premises, a systematic review and meta-analysis of available epidemiological studies were performed to better define and quantify the effect of monotherapy using sulfonylurea relative to metformin monotherapy and risk of cancer in patients with T2DM.

1.4. Research question and objective

The objective of this meta-analysis is to assess the association between cancer and exposure to monotherapy with sulfonylurea versus metformin among subjects with T2DM in a meta-analysis of epidemiological studies. To this end, this meta-analysis will answer the following question:

Research question: Are T2DM patients treated with sulfonylurea at higher risk of cancer incidences compared to T2DM patients who are treated with metformin?

1. METHODOLOGY

2.1. Study Design

This meta-analysis was based on a systematic and comprehensive literature review that was conducted to identify eligible observational studies from peer-reviewed scientific journals. Studies published before April 30, 2017 were identified using the search strategy detailed below.

2.2. Search strategy

Electronic databases were searched including MedLINE/PubMed, and Cochrane Central Register of Controlled Trials (CENTRAL), for published studies and ClinicalTrials.gov for registered trials with the following terms/search detail: "diabetes mellitus, type 2" AND "metformin" AND "sulfonylurea" AND "cancer". The reference lists of the identified articles were manually scanned to identify any other relevant studies. After deleting duplicate articles, the titles and abstracts of search results are assessed and selected relevant studies according to the following inclusion/exclusion criteria.

2.3. Inclusion and Exclusion Criteria

The inclusion criteria were the following:

- Studies on type 2 diabetic patients that reported data on exposure to monotherapy with sulfonylurea, in comparison with metformin, and cancer incidence;
- Studies presenting the relative risk (RR), odd ratio (OR) or hazard ratio (HR) estimation, 95% confidence intervals (CIs) or P-value, size of baseline samples, or other information that could help to interpret the results; and
- Studies written in English language.

The exclusion criteria were the following:

- Studies that involve therapies with antidiabetics other than metformin and sulfonylurea
- Studies that showed no association between metformin or sulfonylurea and cancer;
- Experimental animal studies;
- Review; and

- Absence of relevant data;
- Duplicate articles that have up-to-date versions available
- Studies that failed to publish the full reports

2.4. Data Collection

The details of the studies included in the review were extracted including; study design, country, publication year, sample size, total number of cases and control in sulfonylureas and metformin group, cancer site, mean follow up period, adjustment and stratification variables, event and non-event, RR (or other association measures), and the corresponding 95% CI.

2.5. Statistical Methods

The data were analyzed using Meta-Essentials 1.0 [27]. The summary risk ratio (RR) for exposure to metformin versus sulfonylurea was the measure of interest. Analyses were performed for any site or over all cancer risks. For analysis of these studies, a random effect model was used to pool the raw data, using the Mantel–Haenszel (MH) estimator, and risk ratios (RRs) and corresponding 95% CIs are reported. In addition, a random-effect MH model was used to pool the adjusted estimates reported from the studies. Moreover, the total variation across studies that is due to heterogeneity rather than chance was evaluated using the I^2 statistic [28]. In addition, sensitivity analyses were conducted to examine the robustness of the estimates using an alternative meta-analysis model (random- vs fixed-effect model) and the risk of publication bias was assessed by funnel plots. For all hypothesis tests, evidence was based on $p < 0.05$, and the 95% CIs were therefore presented.

2. RESULTS

3.1. Literature Search

As shown in Figure 1, the initial search yielded 636 articles, of which 44 duplicated ones have deleted. From the 592 remaining papers, 522 ones have excluded based on the title and abstract. The remaining 70 articles were considered of interest, and their full text was retrieved for detailed evaluation. Of these, 60 articles were further excluded because they did not satisfy the inclusion criteria. The remaining 10 studies [22, 25, 29, 30–36] complied with the inclusion criteria and were considered for meta-analysis.

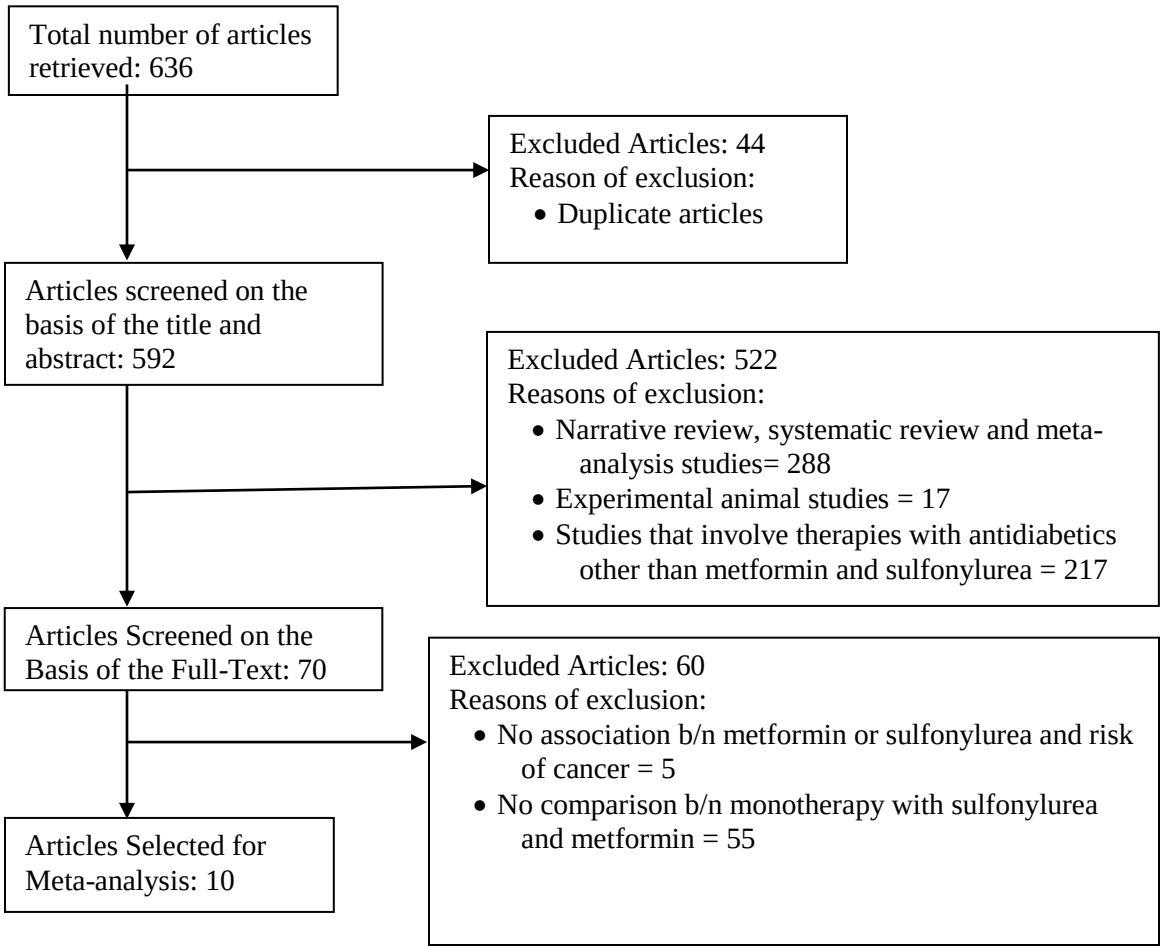

Figure 1: Flow chart of the search result and selection of studies for inclusion in the meta-analysis.

3.2. Study Characteristics

All of the ten studies that met the inclusion criteria were cohort studies. As indicated in Table 1, eight studies were based on European [22, 25, 29, 31, 33, 34, 35, 36], one study was based on USA T2DM patients [32], and the other one study was based on Asians [30]. All the included studies have published between 2009 and 2015.

On the other hand, all the included studies have done a comparison of the risk of cancer associated with monotherapy with sulfonylurea versus monotherapy with metformin in T2DM patients. As shown in the Table 1, the sample sizes of the SUs groups in these studies ranged from 4334 to 32438, with sample sizes of metformin groups ranging from 708 to 132960. The median length of follow-up of treated patients was 3.25 years. Furthermore, the total population involved in the 10 studies were 609,429 (SUs group= 155, 203 and metformin group= 454,226). The total number of cancer cases was 17,164 (SUs groups= 6,987 [4.5%], metformin groups=10,177 [2.2%]).

Table 1: characteristics of included studies

First Author, year, Country, ref	Study design	Cancer site	Sulfonylurea group		Metformin group		Reported RR (Adjusted)	Mean Follow up time (years)	Adjusting variables
			Case	Total (male, %)	Case	Total (male, %)			
Currie CJ, 2009, UK, [29]	Cohort	Any site	479	7,439 (54.9)	1482	31,421(51.1)	1.36(1.23,1.50)	2.4	Smoking, comorbidity, HbA1c, DM duration, weight
Hsieh MC, 2012, Taiwan, [30]	Cohort	Any site	470	6072(53.8)	199	3963(48.3)	1.01(0.85,1.20)	4.2	Age, sex, and duration of DM
Redaniel MT, 2012, UK, [31]	Cohort	Breast	93	4,815(100)	151	11,918(100)	1.02(0.76,1.36)	4.6	Age, region, BMI, year of dx
Ruiter R, 2012, Netherlands, [22]	Cohort	Any site	1962	32591 (48.2)	1590	52698 (46.4)	1.11(1.03,1.20)	3.5	Age, number of other drugs used in the year before the start of OGLD, number of hospitalizations in the year before the start of OGLD, calendar time
Lehman DM, 2012, USA, [32]	Cohort	Prostate	321	4334 (100)	39	708 (100)	1.02(0.74,1.42)	4.8	HbA1c, age, DM duration, & race/ethnicity & medication propensity score, each group
Currie CJ, 2013, UK, [33]		Any site	805	16218 (54.4)	2,581	58,532 (55.9)	1.10(1.02,1.19)	2.48	HbA1c, total cholesterol, serum creatinine, BMI, smoking status, antihypertensive-lipid- lowering, antiplatelet therapy, duration of DM, prior history of cancer, LVD, microvascular disease, number of contacts with the general practitioner in the year prior to the index date, Charlson comorbidity index
Qiu H, 2013, UK, [34]	Cohort	Any site	1165	16904 (59.1)	1389	39070 (57.0)	1.07(0.98,1.17)	3.4	Age, gender; anti-diabetic medication was monotherapy
Mamtani R, 2014, UK,[25]	Cohort	Bladder	66	16128 (55.0)	196	71472 (55.8)	1.04(0.75,1.43)	2	Age, sex, smoking, HbA1c level, obesity
Tsilidis KK, 2014, UK, [35]	Cohort	Any site	1,502	18264 (57.9)	2,303	51484 (56.1)	1.13(1.05,1.22)	2.5	Age, sex, smoking, BMI, alcohol, aspirin, statins, & year of first antidiabetes prescription
Goossens ME, 2015, UK ,[36]	Cohort	Bladder	124	32438 (57.4)	247	132960 (52.1)	1.03(0.78,1.36)	2.6	Age, gender, smoking, BMI, & duration of DM

Abbreviation: N: number, BMI: Body Mass Index, HbA1c: hemoglobin A1C, OGLD: oral glucose lowering drugs

3.3. Meta-analysis

All of the 10 cohort studies reported raw event data. In all, 6,987 (4.5%), cancers were identified from 155, 203 T2DM patients using SUs, compared with 10,177 (2.2%), cancers from 454,226 T2DM patients using metformin. Pooling of those 10 cohort studies suggested that patients using SUs compared with metformin use had a significantly higher risk of overall cancer (RR= 1.59, 95% CI 1.38-1.83, I^2=94.88%, p< 0.00001; Figure 2).

On the other hand, the studies reported adjusted estimates of effects and all these studies adjusted important confounding factors, such as age, sex and duration of DM. After pooling of the adjusted estimates, the use of SUs was not associated with a higher cancer risk than metformin treatment (RR=1.12, 95% CI 1.05–1.19, I^2=51.20%, p< 0.000145; figure 3).

Study	Sulfonylurea		Metformin		Weight (Random, %)	Risk ratio 95% CI
	Cases	Total	Cases	Total		
Currie CJ, 2009	479	7439	1482	31421	10.71%	1.37(1.24,1.51)
Hsieh MC, 2012	470	6072	199	3963	10.13%	1.54(1.31,1.81)
Redaniel MT, 2012	93	4815	151	11918	9.06%	1.52(1.18,1.97)
Ruiter R, 2012	1962	32591	1590	52698	10.95%	2.00(1.87,2.13)
Lehman DM, 2012	321	4334	39	708	7.99%	1.34(0.97,1.86)
Currie CJ, 2013	805	16218	2581	58532	10.88%	1.13(1.04,1.22)
Qiu H, 2013	1165	16904	1389	39070	10.88%	1.94(1.80,2.09)
Mamtani R, 2014	66	16128	196	71472	8.82%	1.49(1.13,1.97)
Tsilidis KK, 2014	1502	18264	2303	51484	10.95%	1.84(1.73,1.96)
Goossens ME, 2015	124	32438	247	132960	9.62%	2.06(1.66,2.55)
Summary RR (95%		155, 203		454,226		1.59(1.38,1.83)
Total	6,987	155203	10,177	454226		
Heterogeneity; Q=175.67, T^2= 0.08, df= 9 (P<.0001), I^2= 94.88						
Test for overall effect: Z= 7.41 (P< 0.00001)						

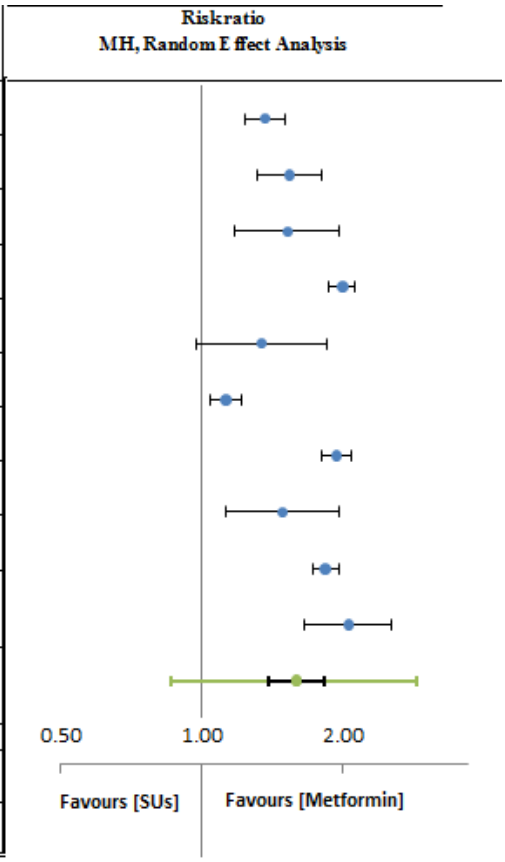

Figure 2: Forest plot of relative risk of cancer when comparing use of sulfonylurea versus metformin, using the raw case data. Blue circles represent study-specific relative risk estimates (size of the blue circle reflects the study-specific statistical weight, that is, the inverse of the variance); horizontal lines represent 95% CIs; gray circle represent summary relative risk estimates with corresponding 95% CIs; *p*-values are from testing for heterogeneity across study-specific raw case data. Abbreviations: CI, confidence interval; RR, relative risk, SU, sulfonylurea.

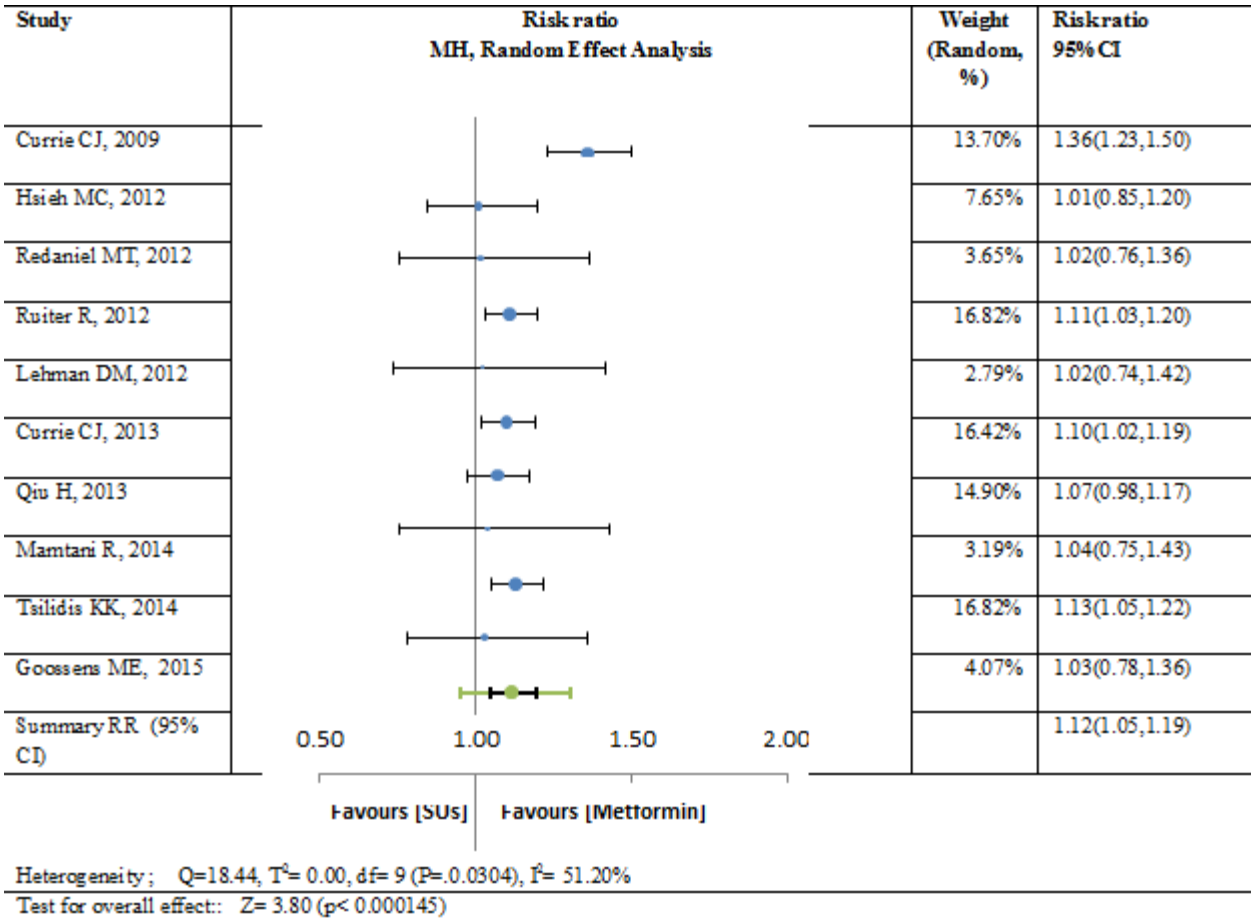

Figure 3: Forest plot of study-specific relative risk estimates (Adjusted) cancer when comparing use of sulfonylurea versus metformin. Blue circles represent study-specific relative risk estimates (size of the blue circle reflects the study-specific statistical weight, that is, the inverse of the variance); horizontal lines represent 95% CIs; gray circle represent summary relative risk estimates with corresponding 95% CIs; *p*-values are from testing for heterogeneity across study-specific estimates. Abbreviations: CI, confidence interval; RR, relative risk, SU, sulfonylurea.

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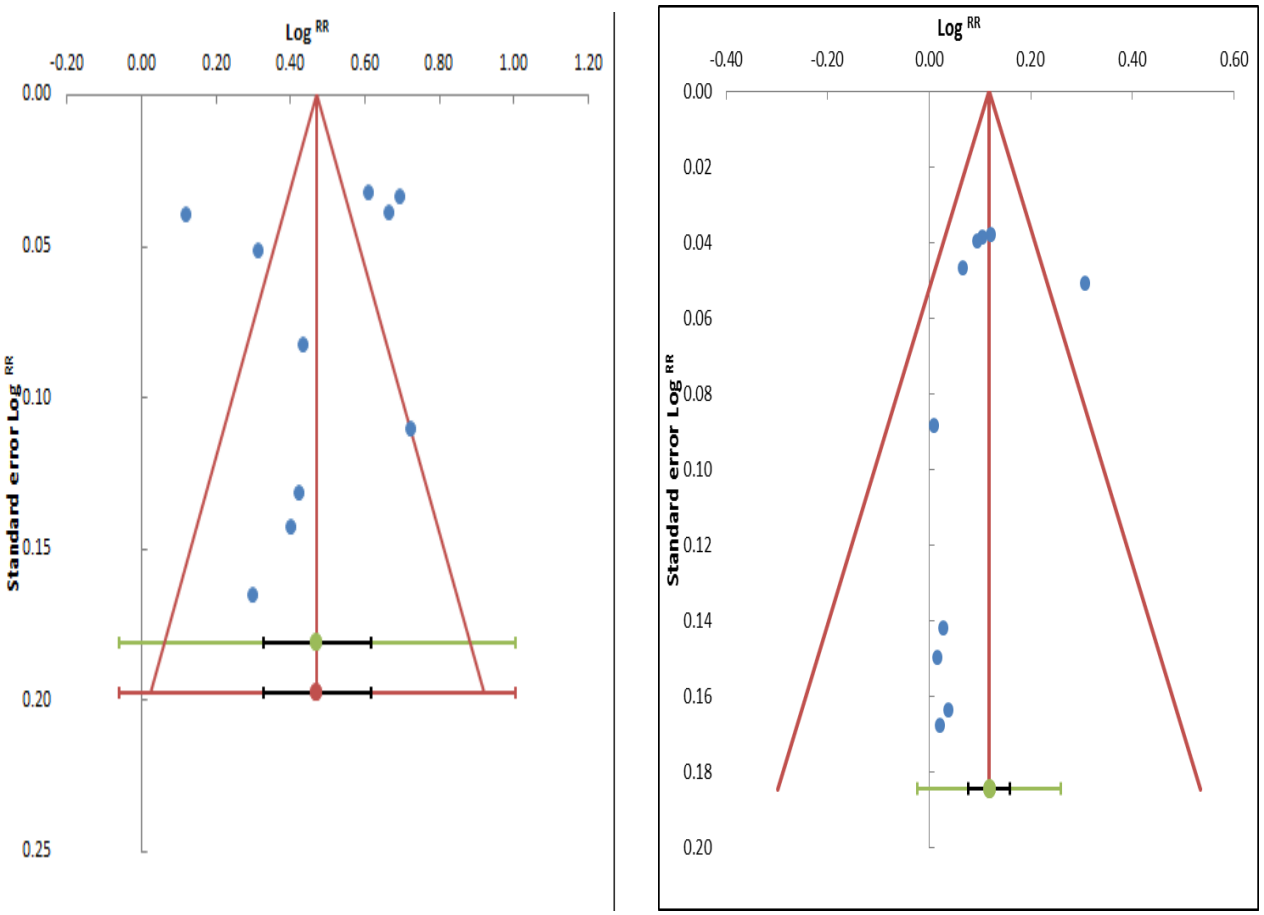
Regarding to heterogeneity and publication bias analysis, when pooling the raw data, there were evidence of large heterogeneity (I^2=94.88%, p= <0.00001; Figure 4), but no evidence for the existence of publication bias (p-value for bias= 0.59). Similarly, in pooling the adjusted RRs, there were moderate evidence of heterogeneity (I^2=51.20%, p< 0.000145; Figure 5), but no evidence for the existence of publication bias (p-value for bias= 0.44).



A. B.

Figure 4: Funnel plot for publication bias in the study investigating cancer risk associated with use of sulfonylurea versus metformin using raw data (A) and using study specific (adjusted) RR estimates. Abbreviations: RR, relative risk; SE, standard error.

3. DISCUSSION

In this study, the results from the analysis of cohort studies suggest that sulfonylurea use significantly increases cancer risk in patients with T2DM compared with metformin. Since sulfonylureas are insulin secretagogues, this observed increase in cancer risk may be a consequence of their effect on insulin levels, and some epidemiological studies link cancer development with the elevated insulin levels observed in patients treated with insulin analogues or insulin secretagogues [18]. However, the exact molecular mechanisms connecting sulfonylurea use to neoplasia are largely unknown. One hypothesis is that sulfonylurea treatment leads to increased insulin and IGF-1 levels and decreased insulin-like growth factor binding protein-1 (IGFBP-1) levels, resulting in increased IGF-1 bioavailability and, in turn, enhanced IGF insulin receptor (IGF-IR) and/or insulin receptor-activation, which ultimately leads to enhanced oncogenesis [12, 13]. Insulin is postulated to promote tumorigenesis both directly and indirectly by acting upon the insulin and IGF-1 receptors expressed on many tumors [37]. Animal studies have also demonstrated that low circulating IGF-1 levels, induced by either caloric restriction or genetic manipulation, are protective against cancer development, while administration of IGFM1 reverses this protective effect [38, 39]. Yet even other studies have shown that hyperglycemia causes a plateau in tumorigenesis and that intensive glucose control does not reduce cancer risk [40, 41].

On the other hand, the current understanding of how sulfonylurea use may impact cancer risk is further complicated by emerging evidence that not all insulin secretagogues appear to promote tumorigenesis. Specifically, in the cohort study by Yang *et al.* [42] both gliclazide (HR=0.67 [95% CI =0.51–0.89]) and glibenclamide (HR=0.65 [95% CI=0.49–0.83]), but not glipizide (HR=1.16 [95% CI=0.84–1.59]), were associated with reduced risk of cancer. Furthermore, in a study by Monani *et al.* [43] gliclazide was actually demonstrated to have an anti-neoplastic effect (OR=0.40 [95% CI=0.21–0.57]). Similarly, gliclazide has also been demonstrated in *in vitro* and *in vivo* animal studies to protect against cancer, mostly through its anti-oxidant effects and by preventing DNA damage [44]. Thus, it may be inappropriate to group the various impacts of different sulfonylureas on cancer risk within one drug class effect, since individual insulin secretagogues may promote dissimilar clinical outcomes.

The major strength of this study is that, it was restricted to data with monotherapy of sulfonylurea versus metformin use on cancer incidence in patients with T2DM. Moreover, the study pooled the RR using both raw case data and study specific (adjusted) RRs to estimate the overall risk. However, one of the limitations of this study is in that, the study was not able to pool the results of other epidemiological studies (RCT and case control studies), because of unavailability of enough studies that investigated the risk of cancer in T2DM patients who received SUs versus metformin. As reported in the result section only one RCT and one case control studies were met the inclusion criteria. Furthermore, some studies have suggested that individual or different generations of sulfonylureas may differentially affect cancer risk [42]. However, this study does not examine for the specific effects of individual sulfonylurea sub-groups or of different sulfonylurea generations on cancer risk, since the small sample sizes of those subgroups would have eliminated the study power to detect meaningful associations.

Furthermore, this study is also limited in that; the cohort studies included in this analysis may also have time-related biases. These biases include immortal time bias, which is introduced with time-fixed cohort analyses that misclassify unexposed time as exposed; time-window bias, which is introduced because of differential exposure opportunity time windows between subjects; and time-lag bias, which is introduced by comparing treatments given at different stages of a disease [45]. The results of the studies included in the current analysis that have these biases may incorrectly indicate that cancer risk is reduced or increased through sulfonylurea or metformin use, respectively, due to misclassification of the exposure time for these medications. However, this is a major limitation of the observational study design and not the methodology used in this meta-analysis.

4. CONCLUSION AND RECOMMENDATION

In conclusion, the current body of evidence clearly shows that compared to metformin SUs can increase cancer risk over the use of metformin. The evidence is not yet adequate to establish the effect of SUs, relative to metformin on cancer. Future investigators should consider more rigorous study design and data analysis, and use data with acceptable rigor.

Finally, it is known that, about 422 million adults were living with diabetes in 2014, and the WHO projects that diabetes deaths will double in 2030 [3]. Moreover, according to the most recent data released by WHO in 2012, more than 14 million of new cancer cases were diagnosed, and 8.2 million cancer deaths and 32.4 million people living with cancer (within 5 years of diagnosis) were registered worldwide [7]. These figures clearly indicate that the disease burdens of both diabetes and cancer are increasing globally, fomenting a health crisis. If medications like sulfonylureas compared with metformin indeed help to reduce the ever-growing economic and health-care burdens of cancer and diabetes, more mechanistic and well-designed long term/large scale cohort studies need are required to provide cost-effective treatment options. As the global need for low cost preventative measures that combat these diseases rises, the exploration of such measures is more tactical and prudent than ever.

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