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Metformin reduces all-cause mortality and diseases of ageing independent of its effect on diabetes control: a systematic review and meta-analysis

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Highlights
• Diabetics on metformin have lower morality than non-diabetics and other diabetics.
• Diabetics on metformin have less cancer than non-diabetics and other diabetics.
• Diabetics on metformin have less cardiovascular disease than other diabetics.
• Metformin appears to extend health and life spans independent of its effect on diabetes.
• Metformin may be able to extend health and lifespans in the general population.

Abstract

This systematic review investigated whether the insulin sensitiser metformin has a geroprotective effect in humans. Pubmed and Embase were searched along with databases of unpublished studies. Eligible research investigated the effect of metformin on all-cause mortality or diseases of ageing relative to non-diabetic populations or diabetics receiving other therapies with adjustment for disease control achieved. Overall, 260 full-texts were reviewed and 53 met the inclusion criteria. Diabetics taking metformin had significantly lower all-cause mortality than non-diabetics (hazard ratio (HR)= 0.93, 95%CI 0.88-0.99), as did diabetics taking metformin compared to diabetics receiving non-metformin therapies (HR= 0.72, 95%CI 0.65-0.80), insulin (HR=0.68, 95%CI 0.63-0.75) or sulphonylurea (HR= 0.80, 95%CI 0.66-0.97). Metformin users also had reduced cancer compared to non-diabetics (rate ratio= 0.94, 95%CI 0.92-0.97) and cardiovascular disease (CVD) compared to diabetics receiving non-metformin therapies (HR= 0.76, 95%CI 0.66-0.87) or insulin (HR= 0.78, 95%CI
Differences in baseline characteristics were observed which had the potential to bias findings, although statistical adjustments were made. The apparent reductions in all-cause mortality and diseases of ageing associated with metformin use suggest that metformin could be extending life and healthspans by acting as a geroprotective agent.

Keywords: metformin; aging; insulin sensitizer; lifespan; longevity; geroprotection

1.0 Introduction

Ageing is characterised by progressive damage at a cellular and organ level as prevention and repair mechanisms begin to break down (Finkel and Holbrook, 2000; Garinis et al., 2008; Mitchell, 2008). Diseases of ageing – such as cardiovascular disease (CVD), kidney failure and cancer – are the result of the accumulation of this damage and ultimately responsible for organisms dying of old age. With the ageing population of the developed world the extension of healthspan – the period that an individual is functional and free of chronic diseases (Barzilai et al., 2016) – has been identified as an important goal both for the minimisation of human suffering and to enable healthcare systems to cope.

One drug which has been subjected to significant research as a geroprotective factor is the insulin sensitiser metformin. Metformin has been shown to extend the lifespans of model organisms (including mice and C. elegans (Anisimov et al., 2011; Anisimov et al., 2010a; Cabreiro et al., 2013)) and a number of potential mechanisms for its effects have been discussed, including decreased insulin and IGF-1 signalling (Liu et al., 2011), inhibition of mTOR (Kickstein et al., 2010; Nair et al., 2014), reducing the levels of reactive oxygen species (ROS)(Batandier et al., 2006; Zheng et al., 2012), lowering inflammation (Saisho, 2015), reducing DNA damage (Algire et al., 2012; Na et al., 2013), and the activation of AMP-activated protein kinase (AMPK)(Zhou et al., 2001). Its effect on AMPK has received particular attention as it models the intracellular mechanisms of caloric restriction (Gillespie et al., 2016; Lee and Min, 2013) – another intervention which has been found to be capable of extending the lifespans of animal models – but which is not feasible for widespread implementation in humans (Roth and Ingram, 2016). This, among other factors, has led to metformin being described as a caloric restriction mimetic (CRM) (Gillespie et al., 2016; Lee and Min, 2013; Roth and Ingram, 2016).

One aspect of metformin which makes it particularly promising for development as a geroprotective agent is that it is already being widely used in humans for a different purpose. As an insulin sensitiser metformin is a first line therapy for diabetes (Nathan et al.,
Through this use its potential for adverse effects and contraindications have been well characterised, and it has been found to have a broad safety profile (Rojas and Gomes, 2013) which would make it significantly quicker and easier to implement as an intervention for ageing than a previously unutilised drug.

Due to the widespread application of metformin for the management of diabetes, a large amount of data has already been collected on its effects on mortality and diseases of ageing. However, extrapolation of this research evidence to the general population is invariably confounded by the fact that the people who are currently receiving metformin have diabetes and are taking it for the treatment thereof. As such, any benefits seen could be due to improvements in diabetes control. Two analytic strategies have been attempted to overcome this limitation and gain some insight as to whether metformin could be used to improve the healthspan of the general population.

The first strategy is the comparison of the outcomes of diabetics taking metformin to the general or non-diabetic population (Bannister et al., 2014). This approach reasons that as diabetics are on average less healthy than non-diabetics, superior outcomes in the metformin/diabetic group should be a consequence of beneficial effects of metformin overcoming their generally worse health status. A weakness of this strategy is that people taking metformin could be observed to have worse or the same outcomes as non-diabetics in the presence of an actual beneficial effect of metformin due to it being masked by the detrimental effects of diabetes. The second strategy is to compare diabetics taking metformin to diabetics managing their diabetes through other means while statistically controlling for the effects of the different therapies on diabetes management (i.e. HbA1c or diabetes related doctor visits (Lin et al., 2015)). This strategy reasons that any difference in outcomes seen after adjusting for the relative effects on disease management can be attributed to metformin’s activity beyond diabetes control, and are therefore generalisable. This strategy’s weaknesses are that statistical adjustment is an imperfect process – it is always possible that residual confounding remains that could cause any differences observed – and that other antidiabetic drugs could cause harms which in comparison would appear to be beneficial effects by metformin.

However, although additional controlled, experimental research will need to be carried out, there exists a wealth of observational data that can offer evidence on whether metformin could be applied as a geroprotective agent in humans. As such, this systematic review and meta-analysis has been undertaken to identify and synthesise all studies where the effects
of metformin on all-cause mortality or diseases of ageing have been compared to the
general or non-diabetic population or to diabetics managing their diabetes through other
means with adjustment for disease control. Numerous systematic reviews have been carried
out which nominally address this review’s outcomes of interest, however those which
investigate the association between metformin and all-cause mortality do so in the context
of severe diseases in addition to diabetes (usually various forms of cancer) (Coyle et al.,
2016; Gash et al., 2017; Li et al., 2017; Meng et al., 2017; Tang et al., 2017; Zhou et al.,
2017a) and their results therefore cannot be generalised, while those that investigate the
incidence of diseases of ageing do so without attempting to adjust for diabetes control (Liu
et al., 2017; Ma et al., 2017; Pladevall et al., 2016; Tang et al., 2017; Zhou et al., 2017b). As
such, the question of whether metformin could act as a geroprotective agent in humans has
yet to be addressed in a systematic review.

2.0 Methods

2.1 Objective

This systematic review aimed to identify and synthesise all research on the effect of
metformin on all-cause mortality and diseases of ageing which had the potential to give
evidence on whether it could be used as a geroprotective factor to extend life and
heathspan in the general population. It followed an a priori protocol pre-registered with
PROSPERO (CRD42016036098).

2.2 Inclusion criteria

2.2.1 Population

Studies on all-cause mortality were eligible for inclusion if they reported on adults with a
minimum age of 40 years or a mean age of 50 years. This age restriction was applied to
increase the proportion of deaths which would be attributable to age and was not applied to
studies on diseases of ageing. Studies were included where metformin was being given to
patients for the treatment of a particular disease (diabetes in all cases although this was not
an inclusion criterion), however where populations were specifically selected to include a
second serious illness (i.e. patients with cancer who were also being treated for diabetes),
studies were not eligible for inclusion.

2.2.2 Exposure
Studies were included where a group of patients had been treated with metformin of any dosage taken on an ongoing basis.

2.2.3 Comparator

Any controls were eligible for inclusion provided they were not receiving metformin. This included members of the general or non-diabetic population as well as people who were receiving other therapies for diabetes. In the latter case, studies were only included if data was adjusted for the effect of diabetes on disease control (i.e. HbA1c levels or number/rate of diabetes related healthcare visits). Where patients entered into studies were using metformin on an ongoing basis their data was included if diabetes control at baseline was adjusted for, as this reflected how well their diabetes was being managed by metformin. However, if patients were enrolled with incident metformin use and the only adjustment for disease control was made at baseline, the study was excluded as this data would not reflect the effect of metformin.

2.2.4 Outcomes

The two primary outcomes were all-cause mortality and the incidence, onset or prevalence of diseases of ageing. Studies on disease specific mortality were not eligible for inclusion. Any diseases of ageing were eligible for inclusion with the exception of diabetes as any effect metformin had on diabetes incidence could more likely be attributed to it directly treating pre-diabetes as an insulin sensitiser than extending healthspan. Diseases ultimately included were cancer (including breast, lung, colorectal, pancreatic, prostate, oesophageal, thyroid, renal, hepatocellular, and head and neck cancer), CVD (including stroke, myocardial infarction, heart failure, coronary heart disease, macrovascular morbidity and ventricular dysfunction), kidney failure, fracture, open angle glaucoma, cognitive impairment, and carpal tunnel syndrome.

2.2.5 Studies

Both experimental and observational studies (including cohort, case-control and analytical cross-sectional studies) were eligible for inclusion in this systematic review. Case report and case series studies were ineligible.

2.3 Search strategy

The search for published literature included Pubmed and Embase, while the search for unpublished studies included the International Clinical Trials Registry Platform, ProQuest
Dissertations and Theses Global, and OpenThesis. No date restrictions were applied, however only English language articles were eligible for inclusion. The search was carried out in March 2016. Full details of the structure and content of each search undertaken are reported in supplementary material 1. Titles and abstracts were screened followed by the retrieval of potentially relevant full-texts. These were then carefully compared to the inclusion criteria to identify eligible studies. The reference lists of included full-texts were then reviewed, however no additional studies that met the inclusion criteria were identified.

2.4 Appraisal and extraction

Duplicate critical appraisal was carried out using the standardised Joanna Briggs Institute (JBI) appraisal checklists for experimental, cohort, case control and cross-sectional studies (The Joanna Briggs Institute, 2014) by independent reviewers working separately, with consensus reached through discussion in all cases. Data was extracted for all studies using an extraction template which included fields for: study methodology, study design, inclusion criteria, data source, country, disease control, exposure, comparator, sample size/events, follow up, outcome(s), outcome data, and statistical adjustments. Where additional data or clarification was needed, attempts were made to contact corresponding authors by email.

2.5 Data synthesis

Where possible, data was pooled through meta-analysis. The inverse variance method with a random effects model from RevMan was used, unless otherwise specified. Data included in the meta-analyses was always from the analyses with the greatest adjustment. Where summary measures differed, outcomes were converted to hazard ratios (HRs) using the equations described in Tierney et al. 2007 (Tierney et al., 2007). Data were analysed separately based on comparators utilised, and heterogeneity was assessed using the Chi² and I² tests. Two sets of sensitivity analyses were carried out where poor quality studies (<70% on critical appraisal) were removed from meta-analyses, and where studies that did not adjust for duration of diabetes and/or comorbidities were removed. Analysis of publication bias through Funnel plots and Egger’s test was planned if sufficient studies (at least 10) were found for any outcomes, however none were. Where statistical pooling was not possible data is presented narratively.
3.0 Results

The initial search identified a total of 21,027 studies, which was reduced to 19,408 following the removal of duplicates. Following title/abstract screening, 260 studies were included for full-text review. Of these, 207 were excluded and 53 were included. Full search details and reasons for exclusion are given in Fig. 1. Overall, 13 included studies reported data on all-cause mortality while 49 reported on diseases of ageing.

3.1 All-cause Mortality

Of the studies that investigated the effect of metformin on all-cause mortality, four compared diabetic patients being treated with metformin to the general population or non-diabetic patients (Bannister et al., 2014; Berard et al., 2011; Bo et al., 2012; Claesen et al., 2016), while nine compared diabetic patients being treated with metformin to diabetic patients managing their diabetes through other means. Subgroups within the latter group included insulin therapy (Ekstrom et al., 2012; Ghotbi et al., 2013), diet interventions (Bo et al., 2012; Sullivan et al., 2011), sulphonylurea therapy (Evans et al., 2006; Kahler et al., 2007; Sullivan et al., 2011; Wang et al., 2014), and non-metformin therapies (studies assigned to this group included all patients receiving any other oral hypoglycemic agent, but may have also included those receiving insulin, diet, or no therapy as well) (Ekstrom et al., 2012; Ghotbi et al., 2013; Gosmanova et al., 2008; Libby et al., 2009; Pratipanawatr et al., 2010).

Full details of interventions, controls and other study characteristics are included in table 1. For the metformin versus non-diabetic comparison three of the cohorts were matched by age (Bannister et al., 2014; Bo et al., 2012; Claesen et al., 2016) while in the fourth (Berard et al., 2011) metformin users were older (mean±SD age 56.2±6.6) than non-diabetics (50.1±8.3); a disparity which could be expected to bias the results towards a finding of metformin being associated with increased mortality, although age was adjusted for. In the majority of cases when metformin users were compared to other diabetics, metformin users were younger (Ekstrom et al., 2012; Evans et al., 2006; Ghotbi et al., 2013; Gosmanova et al., 2008; Kahler et al., 2007; Sullivan et al., 2011; Wang et al., 2014). Differences between mean ages ranged from 0.4 years (Gosmanova et al., 2008) to 5.9 years (Ekstrom et al., 2012). In one study the metformin group was older than the control by <1 year (Bo et al., 2012), and in two mean ages were not reported (Libby et al., 2009; Pratipanawatr et al., 2010), although Libby et al. did match by year of diagnosis so age is likely to have been close. The trend of metformin users being younger than other diabetics has the potential to bias results towards a finding of decreased mortality, however all studies adjusted for age, with the exception of...
Bo et al. The appraisal of study quality found that overall the studies were conducted rigorously, with notable deficiencies being that none reported similar characteristics in their respective populations at baseline and less than half had adequate follow up times for our observations of interest (defined as ≥5 years) with thorough reporting on reasons for loss to follow up (Supplementary material 2).
Meta-analysis of the four studies that compared people managing their diabetes with metformin to non-diabetics or the general population (Bannister et al., 2014; Berard et al., 2011; Bo et al., 2012; Claesen et al., 2016) found that people taking metformin had significantly lower mortality compared to those who were not (Fig. 2 HR= 0.93, 95%CI 0.88 to 0.99, p=0.03). One included study was an outlier in that it reported a non-significant increase in mortality for people taking metformin (Berard et al., 2011), however it was by far the smallest study, with just nine events in the metformin group. Hazard ratios (HRs) used in the meta-analysis represent adjusted values with two exceptions where the HR could only be calculated for crude data (Bannister et al., 2014) and where only crude analyses were performed for this data (Bo et al., 2012). Sensitivity analysis could not be carried out as no studies were of poor quality and only one study adjusted for diabetes duration and/or comorbidity.

The finding that metformin reduces mortality was supported by analyses that compared diabetic patients receiving metformin to diabetic patients receiving other therapies (Fig. 3), where diabetes control had been adjusted for.

The majority of subgroups, including studies with non-metformin controls (HR= 0.72, 95%CI 0.65 to 0.80, p<0.00001), insulin treated controls (HR=0.68, 95%CI 0.63 to 0.75, p<0.00001), and sulphonylurea treated controls (HR= 0.80, 95%CI 0.66 to 0.97, p= 0.02), found that even after adjusting for metformin’s effect on diabetes control, diabetics taking it had lower all-cause mortality than diabetics who were not. The one exception was the subgroup where the controls managed their diabetes through diet alone. In this case one study (Bo et al., 2012) found a significant benefit to all-cause mortality for metformin and the other study (Sullivan et al., 2011) found a non-significant disadvantage resulting in an overall non-significant finding (HR=0.91, 95%CI 0.68 to 1.22, p= 0.53). An overall meta-analysis could not be performed as the same studies were included across multiple subgroups where they
presented their metformin data relative to different controls. However, when the duplicate data sets were excluded (removing those with the strongest positive findings: Ekstrom Non-Metformin, Ghotbi Non-Metformin and Sullivan Sulphonylurea), diabetics taking metformin had significantly lower mortality compared to controls after adjusting for disease control (HR= 0.75, 95%CI 0.69 to 0.82, p<0.00001). Sensitivity analysis for the non-metformin control subgroup where two studies which did not adjust for duration of diabetes and/or comorbidity were excluded (Gosmanova et al., 2008; Libby et al., 2009) did not affect results. Sensitivity analysis for study quality could not be performed as no studies were poor quality.

### 3.2 Diseases of ageing

The main diseases of ageing investigated in the included studies were cancer (Supp table 1) and cardiovascular disease (CVD, Supp table 2). However, other age related conditions were investigated in a smaller number of studies including renal failure, fracture, open angle glaucoma, cognitive impairment and carpal tunnel syndrome (Supp table 3).

#### 3.2.1 Cancer

Overall, 25 studies investigated the association between metformin use and cancer (Supp table 1). Where people taking metformin were compared to the general or non-diabetic population the majority of studies did not report the ages of these groups. The two that did found that people taking metformin were significantly older (Andersson et al., 2012; But et al., 2014), potentially biasing the results to increased risk of cancer in metformin users although age was adjusted for in both analyses. Amongst the remainder of studies half were case-controls which matched for age and are therefore unlikely to have meaningful hidden bias (Becker et al., 2014, 2016; Bodmer et al., 2012a; Bodmer et al., 2012b; Lu et al., 2015; Walker et al., 2015) and the remainder adjusted for age (Baur et al., 2011; Becker et al., 2013; Nordström et al., 2015; Tseng, 2011, 2012a, b). For people taking metformin compared to diabetics controlling their diabetes through other means, only one study reported relative ages for the comparison (metformin users were younger biasing results towards a lower risk of cancer although age was adjusted for (Goossens et al., 2015)). Nine studies were case-controls which matched for age and therefore have limited risk of bias in this regard (Azoulay et al., 2011; Becker et al., 2013; Bodmer et al., 2010; Bosco et al., 2011; Chen et al., 2013; Mazzone et al., 2012; Sehdev et al., 2015; Smiechowski et al., 2013a;
Smiechowski et al., 2013b), while two cohort studies adjusted for age but did not report it (Libby et al., 2009; Redaniel et al., 2012). Critical appraisal showed that cohort studies compared groups with significant differences at baseline, tended to lack adequate follow up and did not report on reasons for loss to follow up with similar deficiencies observed in case-control studies (supplementary material 2). Of the included studies, three investigated the incidence of any cancer for people taking metformin for diabetes control compared to non-diabetics. Two of these studies could be combined in meta-analysis (Fig. 4A, (Andersson et al., 2012; But et al., 2014)) and showed that diabetics taking metformin had significantly lower cancer incidence (Rate ratio= 0.94, 95%CI 0.92 to 0.97, p=0.0003). In this case rate ratio was used in the meta-analysis rather than HR as both studies used it as the summary measure to report their data. It should be noted that although there was essentially no heterogeneity between the two studies, the results of the meta-analysis drew almost entirely on the findings of (Andersson et al., 2012) which received 99% of the weight. The third study (Baur et al., 2011) was cross-sectional and found a non-significant decrease in the prevalence of cancer in diabetics receiving metformin monotherapy compared to the non-diabetic population, and a non-significant increase in diabetics receiving metformin as a combination therapy. A fourth study investigated the effect of metformin on cancer incidence but compared it to matched diabetics receiving any other oral hypoglycaemic agent rather than non-diabetics (Libby et al., 2009). It reported a significant reduction for metformin (HR= 0.63, 95%CI 0.53 to 0.75, p<0.05), supporting the findings of the meta-analysis.

Three studies investigated the effect of taking metformin on the development of pancreatic cancer in people with diabetes compared to the general (Bodmer et al., 2012b; Lu et al., 2015) or non-diabetic population (Walker et al., 2015). However, findings were mixed and the result of the meta-analysis was non-significant (Fig. 4B, all studies reported their outcomes as odds ratios (ORs), therefore this was the summary measure used in the meta-analysis). Sensitivity analysis could not be carried out as no studies were of poor quality and only one study adjusted for duration of diabetes and/or comorbidity.
The effect of metformin on the incidence of colorectal cancer was investigated in four studies, three of which were similar enough to be combined in meta-analysis (Libby et al., 2009; Sehdev et al., 2015; Smiechowski et al., 2013a). These studies compared the incidence of colorectal cancer in diabetic patients taking metformin to non-metformin diabetic controls. Although a trend for metformin use being associated with a reduced incidence of colorectal cancer was found, the results were ultimately non-significant (Fig. 5A, HR= 0.85, 95%CI 0.72 to 1.02, p=0.08). As heterogeneity was non-significant (Chi²= 0.15, I²= 48%) and all studies suggested a benefit, a meta-analysis using the fixed effects model was trialled. A significant reduction in colorectal cancer incidence for the metformin group was found (HR= 0.91, 95%CI 0.84 to 0.98, p=0.009). Sensitivity analysis where one study which did not adjust for duration of diabetes and/or comorbidity was excluded (Libby et al., 2009) did not have a large effect on the estimate but resulted in a significant finding by the random effects model (HR= 0.92, 95%CI 0.85 to 0.99, p=0.02). Sensitivity analysis for study quality could not be performed as no studies were poor quality. An additional study not included in the meta-analysis (Tseng, 2012a) compared the incidence of colorectal cancer in diabetics taking metformin to the general population and found that metformin was associated with a significant reduction (relative risk= 0.73, 95%CI 0.58 to 0.92, p=0.008), supporting the conclusion that metformin may reduce colorectal cancer incidence.

Breast cancer incidence was investigated in four studies, three of which compared the incidence in diabetic patients taking metformin to diabetic patients who were not taking metformin (Bodmer et al., 2010; Bosco et al., 2011; Libby et al., 2009). These were combined in meta-analysis Fig. 5B which showed a significant reduction in the incidence of breast cancer (HR= 0.71, 95%CI 0.54 to 0.92, p=0.01). Sensitivity analysis where one study which did not adjust for duration of diabetes and/or comorbidity was excluded (Libby et al., 2009) did not have a large effect on the estimate but resulted in a non-significant finding being made (HR= 0.72, 95%CI 0.50 to 1.04, p=0.08). Sensitivity analysis for study quality could not be performed as no studies were poor quality. The fourth study compared the incidence of breast cancer in diabetics being treated with metformin to diabetics being treated with sulphonylurea (Redaniel et al., 2012) and found a non-significant increase in breast cancer for metformin. Interestingly, they also compared people taking metformin in combination with sulphonylurea to people taking sulphonylurea alone and found a significant reduction in the incidence of breast cancer in the people treated with metformin (HR= 0.66, 95%CI 0.50 to 0.88, p<0.05).
The effect of metformin on the incidence of lung cancer was investigated in four studies. Three studies compared the incidence of lung cancer in diabetic patients treated with metformin to diabetic patients not treated with metformin (Libby et al., 2009; Mazzone et al., 2012; Smiechowski et al., 2013b) and could be combined in meta-analysis Fig. 5C. The combined results showed that taking metformin was associated with a decreased incidence of lung cancer in diabetic patients (HR= 0.80, 95%CI 0.65 to 0.98, p=0.03). Sensitivity analysis could not be carried out as no studies were of poor quality and only one study adjusted for duration of diabetes and/or comorbidity. The fourth study (Bodmer et al., 2012a) compared diabetics taking metformin to the general population. It did not find any significant effects for short term (1-14 prescriptions) or long term (≥40 prescription) metformin use, however medium term (15-39 prescriptions) metformin use was contradictorily associated with a significant increase in the odds of developing lung cancer (OR= 1.24, 95%CI 1.03 to 1.50, p<0.05).

Additional cancers that were investigated included prostate, bladder, thyroid, renal, head and neck, oesophageal, and hepatocellular cancer. Neither of the two studies on prostate cancer found any significant association with metformin use (Azoulay et al., 2011; Nordström et al., 2015). Nor did the studies on bladder cancer (Goossens et al., 2015; Tseng, 2011), thyroid cancer (Tseng, 2012b), renal cell carcinoma (Becker et al., 2016), head and neck cancer (Becker et al., 2014), or oesophageal cancer (Becker et al., 2013). However, metformin users had significantly lower odds of developing hepatocellular cancer compared to other diabetics (OR= 0.79, 95%CI 0.75 to 0.85, p<0.0001).

### 3.2.2 Cardiovascular disease

Fifteen studies were found that investigated the effect of metformin on the development of CVDs (Supp table 2). Critical appraisal showed a similar pattern in quality as observed for studies on all-cause mortality and cancer – groups had significant differences at baseline, follow-up was frequently shorter than desirable to observe the onset of disease, and loss to follow up was inadequately described (supplementary material 2). Additionally, over half of
the cohort studies included participants who already had some form of CVD at baseline and half of the case-control studies did not adequately control for potential confounding factors.

No studies compared people taking metformin for diabetes management to non-diabetics or the general population. For those that compared people taking metformin to people controlling their diabetes through other means, six included analyses where the group receiving metformin was younger than those who were not (Ekstrom et al., 2012; Evans et al., 2006; Ghotbi et al., 2013; McAlister et al., 2008; Sullivan et al., 2011) (the difference ranged from 1.1 years (Sullivan et al., 2011) to 5.9 years (Ekstrom et al., 2012), while in two studies people receiving metformin were younger than comparators (2.8 years (Nichols et al., 2005) and 5 years (Kooy et al., 2009)). Three further studies were case-controls which matched for age (Floyd et al., 2016; Hartung et al., 2005; Koro et al., 2005) while four did not report on relative ages but did adjust for age in their analyses (Gejl et al., 2015; Giorda et al., 2011; Jansson et al., 2014; Peters et al., 2013). Five studies comparing the incidence of any CVD in diabetics taking metformin to diabetics not receiving metformin could be combined in meta-analysis (Ekstrom et al., 2012; Gejl et al., 2015; Ghotbi et al., 2013; Jansson et al., 2014; Peters et al., 2013). This showed a significant reduction in CVD for people taking metformin (Fig. 6, HR= 0.76, 95%CI 0.66 to 0.87, p<0.0001). Sensitivity analysis where studies of low quality were excluded from meta-analysis did not have a large impact on the estimate (HR= 0.73, 95%CI 0.53 to 1.00, p=0.05). No studies did not adjust for duration of diabetes and/or comorbidity. The finding of a reduction was supported by two studies that compared the incidence of any CVD between diabetics being treated with metformin to diabetics treated with insulin (Ekstrom et al., 2012; Ghotbi et al., 2013), where a significant decrease in incidence with metformin use was found (Fig. 6, HR= 0.78, 95%CI 0.73 to 0.83, p<0.00001). However, the meta-analysis for sulphonylurea controls, which also included two studies (Evans et al., 2006; Sullivan et al., 2011), did not show a significant effect (Fig. 6). One study was found that investigated the effect of metformin on incidence of CVD compared to diabetics managing their diabetes through diet and showed a non-significant increase in the metformin group (Sullivan et al., 2011). An overall meta-analysis was prevented as the subgroups contained data from the same participants. However, when duplications were removed (again excluding the data that showed the strongest positive effect: (Ekstrom Non-metformin, Ghotbi Non Metformin, Sullivan Diet), the result was a significant effect for metformin reducing the incidence of CVD (HR= 0.83, 95%CI 0.73 to 0.94, p=0.004).
Three studies investigated the effect of metformin on the incidence of stroke. Two compared the incidence of stroke in metformin users to other diabetics not using metformin (Floyd et al., 2016; Jansson et al., 2014). Meta-analysis of their findings (Fig. 7A) showed a significant reduction in stroke for metformin users (HR= 0.70, 95%CI 0.53 to 0.93, p=0.01). The third study (Sullivan et al., 2011) compared metformin use to diabetics managing their diabetes through diet alone or with sulphonylurea, and found a non-significant increase in the incidence of stroke with metformin use in both cases.

Myocardial infarction incidence was examined in just two studies (Floyd et al., 2016; Jansson et al., 2014) and although (Jansson et al., 2014) showed a strong significant effect in favour of metformin, the result of the meta-analysis was a non-significant decrease in myocardial infarction for metformin users compared to diabetics not using metformin (Fig. 7B, HR= 0.63, 95%CI 0.28 to 1.42, p=0.27).

Heart failure was investigated in four studies, however due to differences in study design and outcomes reported no meta-analysis could be performed. One study (Hartung et al., 2005) compared metformin users to other oral hypoglycaemic agent users with regards to hospitalisations resulting from heart failure, but found no significant effect. Another study (Koro et al., 2005) found non-significant increases for metformin on incidence of congestive heart failure compared to diabetics with no drug exposure and diabetics treated with sulphonylurea. Another study (Nichols et al., 2005) showed a non-significant decrease in congestive heart failure for diabetics treated with metformin compared to those treated with sulphonylurea. It also found a significant decrease in heart failure with metformin compared to insulin use (rate ratio= 0.39, 95%CI 0.19 to 0.80, p<0.05). The final study (McAlister et al., 2008) compared the incidence of heart failure in metformin treated patients to those treated with sulphonylurea and found a non-significant decrease in heart failure for those receiving metformin.

Results for coronary heart disease also could not be meta-analysed. One study (Peters et al., 2013) found a non-significant reduction for users of metformin compared to other diabetics who were not receiving metformin, while another (Sullivan et al., 2011) found significant
increases with metformin use compared to patients being treated with diet alone or sulphonylurea monotherapy. Other CVD outcomes investigated were left ventricular dysfunction, which was significantly more prevalent in diabetics being treated with metformin compared to those not taking metformin (OR=1.62, 95%CI 1.09 to 2.40, p=0.16) – although the authors of this cross-sectional study concluded that this outcome was likely due to confounding due to numerous large between group differences (Giorda et al., 2011) – and macrovascular morbidity and mortality (Kooy et al., 2009) which was significantly lowered by the addition of metformin to insulin therapy compared to the addition of a placebo to insulin therapy (HR=0.34, 95%CI 0.21 to 0.56, p=0.001).

3.2.3 Other diseases of ageing
Seven studies investigated the effect of metformin on diseases of ageing, other than cancer and CVD, although none could be combined in meta-analysis (Supp table 3). Critical appraisal of studies (supplementary material 2) showed differences between groups at baseline as well as short follow up with no reasons given for losses. Two studies compared outcomes for people using metformin to the general or non-diabetic population; neither reported on differences in ages, although one was a case-control study which matched for age (Geoghegan et al., 2004) and the other adjusted for age (Vestergaard et al., 2005). Amongst the five studies which compared people receiving metformin to those controlling their diabetes through other means there was heterogeneity in reporting; two studies included a cohort of metformin patients who were younger than their comparators (Hung et al., 2013; Masica et al., 2013), one reported that one subgroup of metformin users was younger and another older but did not report the overall comparison (Ng et al., 2014), one reported that metformin users were slightly older than patients assigned to a different medication (in an RCT design) (Kahn et al., 2008), and one did not report relative ages (Lin et al., 2015). All studies adjusted for age in their analyses with the exception of the RCT. Two studies investigated kidney failure or decline. One study (Hung et al., 2013) found a non-significant decrease for the incidence of a glomerular filtration event or end stage renal disease for diabetic patients treated with metformin compared to diabetic patients treated with sulphonylurea, however this decrease was significant in a subgroup of patients where urine protein at baseline was known and adjusted for (HR=0.78, 95%CI 0.64 to 0.97). The same analyses were performed for patients treated with metformin in addition to sulphonylurea compared to sulphonylurea alone, however no significant differences were found. The second study found a non-significant reduction in the decline of estimated glomerular
filtration rate to <60ml/min/1.73m² within one year for metformin users (Masica et al., 2013) compared to sulphonylurea users, however there was a significant reduction in risk of developing proteinuria (HR= 0.71, 95%CI 0.53 to 0.95, p<0.05). These comparisons were repeated for treatment with thiazolidinediones compared to metformin, and no significant differences were found.

The effect of metformin on fracture risk was investigated in two studies. One study (Kahn et al., 2008) showed that metformin significantly decreased risk of fracture relative to treatment with rosiglitazone (HR= 0.64, 95%CI 0.46 to 0.88, p=0.007). Stratified analysis showed that the effect persisted in women but not in men. The other study (Vestergaard et al., 2005) found the metformin users had lower risks of fracture compared to the general population for <150 defined daily doses (DDD) (OR=0.87, 95%CI 0.86 to 0.96, p<0.05), 150-499 DDD (OR= 0.81, 95%CI 0.71 to 0.94, p<0.05), and ≥500 DDD (OR= 0.81, 95%CI 0.70 to 0.93, p<0.05) consumed from the study start date to the date of censoring.

Additional studies on diseases of ageing investigated open angle glaucoma (Lin et al., 2015), which had a significantly lower incidence in diabetics taking metformin compared to other diabetics (HR= 0.75, 95%CI 0.59 to 0.95, p<0.05), cognitive impairment (Ng et al., 2014), which was significantly less likely to develop in patients taking metformin compared to other diabetics (OR= 0.49, 95%CI 0.25 to 0.95, p<0.05; a finding which remained significant in users who took metformin for more than 6 years before baseline (OR= 0.27, 95%CI 0.12 to 0.60, p<0.05), but not those who took it for 6 years or less), and carpal tunnel syndrome (Geoghegan et al., 2004), which was not significantly associated with metformin use relative to the general population.

4.0 Discussion

This systematic review has shown through meta-analysis that diabetics taking metformin have a lower rate of all-cause mortality than non-diabetic people and the general population. Our results suggest that metformin could be an effective intervention to extend the lifespans of people who do not have diabetes. This is supported by additional meta-analyses showing that diabetics taking metformin had lower rates of all-cause mortality than
diabetics receiving other therapies after adjusting for relative disease control. This was true for metformin compared to diabetics receiving any non-metformin therapy, insulin, or sulphonylurea, however, the result was non-significant for diet therapy. This finding could be due to diet therapy being a more likely option for people with early or less severe diabetes, or it could alternatively be explained by improved diet and lifestyle having an effect beyond disease control comparable to metformin. With only two studies contributing to the meta-analysis further research is needed for conclusions to be drawn.

Other results suggest that metformin’s effect on all-cause mortality could be due to it having a geroprotective effect of delaying or preventing diseases of ageing, particularly cancer and CVD which are two of the leading causes of death and disability worldwide (Mathers and Loncar, 2006). We show that diabetic people taking metformin had a lower rate of developing any cancer compared with the general population, and had a lower risk of developing colorectal, breast or lung cancer compared with diabetics managing their diabetes through non-metformin therapies after adjusting for disease control. The rate of any form of CVD was similarly lower for diabetic people taking metformin compared to those managing their diabetes through any non-metformin therapy or insulin therapy after adjusting for disease control, however findings for sulphonylurea were non-significant. The incidence of stroke was also reduced with metformin use compared to any non-metformin therapy; however there was no significant effect for metformin on myocardial infarction.

These findings rest on the credibility of two assumptions: that improvements in survival and reduced onset of diseases of ageing seen in diabetics above the outcomes of people who do not have diabetes can be generalised to the non-diabetic population, and that benefits seen between populations receiving different therapies for diabetes (which are still present after adjusting for disease control) are independent on the therapies’ effects on diabetes and are likewise generalisable. Of the two, the former is the more robust assumption, although the potential for confounding, as with any observational research, does exist. As such, the findings for all-cause mortality and cancer – which are backed by comparisons to the non-diabetic population – should be considered more reliable than those for CVD – which are based solely on comparisons to diabetics treated with other therapies. However, taken together these results provide strong evidence, from a combined population of 417,316 for all-cause mortality compared to the non-diabetic or general population alone, to support the
hypothesis that metformin has a geroprotective effect in humans, slowing the progression of
diseases of ageing and in doing so extending both health and lifespans.

Beyond the assumptions discussed, limitations of this research include that none of the
meta-analyses included sufficient studies for any investigation of publication bias, study
designs were entirely observational (with two exceptions that showed benefits for
metformin on fracture (Kahn et al., 2008) and macrovascular morbidity and mortality (Kooy
et al., 2009)), and none of the data for diseases of ageing beyond cancer and CVD could be
combined through meta-analysis despite promising findings for kidney failure, fracture risk,
open angle glaucoma and cognitive impairment. With regards to cognitive impairment, in
the conduct of this review’s search, numerous additional studies were found which
investigated the effect of metformin on measures of dementia. However, these studies were
not eligible for inclusion due to investigating average level of cognitive ability (rather than
the incidence or prevalence of impairment) or not adjusting for disease control. An
additional systematic review with broader inclusion criteria relating to dementia outcomes is
now planned (Campbell et al., 2017). Diabetes, despite being a major chronic disease which
becomes progressively more common with age, was not investigated by this review as
metformin being an intervention for diabetes was too much of a complicating factor.
However, experimental studies have directly investigated the effect of prophylactic
metformin for the prevention of diabetes (in high BMI populations) and shown that it
significantly reduced its development (Andreadis et al., 2009; Knowler et al., 2009).

The primary issue with study quality in this systematic review, highlighted by critical
appraisal, was that studies rarely compared groups that did not have meaningful differences
at the beginning of the observation period. Where diabetics receiving metformin were
compared to general or non-diabetic populations, this aspect is essentially a feature of the
study design. However, for metformin compared to other diabetic therapies it is more
concerning, as it reflects a trend for people receiving metformin to be younger with shorter
durations of disease, which has the potential to bias results towards finding a protective
effect for metformin. Critical appraisal also showed that studies were generally thorough in
adjusting for potential confounding factors, however, and a sizeable proportion of studies
utilised matching. As such, the potential impact of this source of bias is at least mitigated.
Additionally, the general pattern of findings for metformin compared to other diabetic
therapies being reproduced by findings for metformin compared to the general or non-diabetic population suggests that they are real and not the result of confounding. Studies frequently had follow up periods less than five years, which affects the likelihood of age related outcomes having an opportunity to occur, however the use of summary measures which incorporate time (i.e. hazard and rate ratios) will have accommodated this. Finally, studies did not report on reasons for (or the existence of) loss to follow up which, if known, may have revealed or precluded potential sources of bias.

Future research in this area should include controlled trials. Two existing trials on the effects of metformin on ageing – Metformin in Longevity Study (MILES NCT02432287) and Targeting Ageing with Metformin (TAME) (Barzilai et al., 2016) – are currently planned. The findings of this systematic review suggest that studies on the effect of metformin in older people on the incidence of chronic diseases (healthspan) and longevity (lifespan) over the long term are warranted. It must be stressed, however, that any immediate use of metformin as an intervention for ageing by the general population is not supported. Despite being a relatively safe drug, with feared effects on lactic acidosis and kidney function emerging to be minor (Ekstrom et al., 2012; Vasisht et al., 2010), metformin is not risk free. Although studies in mice have generally been positive, some have shown increased mortality when taken at higher concentrations (Martin-Montalvo et al., 2013), and others have suggested that its effects may be gender exclusive (favouring females (Anisimov et al., 2010b)), an observation which is supported by some subgroup analyses performed in included studies (Bodmer et al., 2012b; Kahn et al., 2008). As such, there is a risk that people taking metformin as a geroprotective agent will experience harm or no effect even if it is ultimately found to be beneficial in some circumstances. Additionally, recent work by Wang et al. – who showed in an included study that the presence of frailty significantly attenuated the effect of metformin on mortality (Wang et al., 2014) – found that older men’s propensity for diseases of ageing significantly interacted with metformin’s effect on the development of these diseases (Wang et al., 2017).

The findings reported in this systematic review remain preliminary generalisations, primarily making use of existing observational evidence collected for other purposes to investigate the credibility of the hypothesis that the insulin sensitiser metformin may extend the health and lifespans of people from the non-diabetic population. Differences in baseline characteristics
were found which had the potential to bias results both towards positive findings (where metformin users were compared to other diabetics) as well as away from (where metformin users were compared to non-diabetics). While they should not be overstated, the apparent association with reductions in all-cause mortality and diseases of ageing found through meta-analysis do support this hypothesis, and metformin should be investigated as an intervention for ageing in future clinical trials.

**Funding**

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

**Figure legends**

Figure 1. PRISMA flow chart.

Figure 2. All-cause mortality hazard ratios in people using metformin for diabetes compared to the general or non-diabetic population.

Figure 3. All-cause mortality hazard ratios in people using metformin for diabetes compared people using other therapies for diabetes. 2.1.2 ‘All Non-Metformin Control’ includes studies where the controls were any diabetic people receiving who were not being treated with metformin (or controls which were receiving a broad range of therapies). 2.1.3 ‘Diet Control’ includes studies where controls were managing their diabetes though diet. 2.1.4 ‘Insulin Control’ includes studies were controls were managing their diabetes using insulin. 2.1.5 ‘Sulphonylurea’ includes studies where controls were managing their diabetes using sulphonylurea.

Figure 4. A) Cancer incidence in people with diabetes taking metformin compared to the general or non-diabetic population. B) Pancreatic cancer incidence in people with diabetes taking metformin compared to the general or non-diabetic population.

Figure 5. A) Colorectal cancer incidence for people with diabetes taking metformin compared to diabetics not taking metformin. B) Breast cancer incidence for people with
diabetes taking metformin compared to diabetics not taking metformin C) Lung cancer incidence for people with diabetes taking metformin compared to diabetics not taking metformin.

Figure 6. Incidence of any CVD in diabetic patients treated with metformin compared to diabetic controls receiving other therapies. 2.5.1 ‘All Non-Metformin Control’ includes studies where the controls were any diabetic patients who were not being treated with metformin (or controls who were receiving a broad range of therapies). 2.5.2 ‘Diet Control’ includes studies where controls were managing their diabetes through diet. 2.5.3 ‘Insulin Control’ includes studies where controls were managing their diabetes using insulin 2.5.4 ‘Sulphonylurea control’ includes studies where controls were managing their diabetes using sulphonylurea.

Figure 7. A) Stroke incidence for diabetics taking metformin compared to diabetics not taking metformin. B) Myocardial infarction incidence for diabetics taking metformin compared to diabetics not taking metformin.

References


outcomes in overweight and obese subjects with type 2 diabetes: a substudy of the SCOUT trial. Diabetes Care 36, 3746-3753.


Figure Caption
Figure 1. PRISMA flow chart.

Papers found by search strategy
n=21,027

Duplicates
n=1,619

Title/Abstract review
n=19,408

Excluded by Title/Abstract review
n=19,148

Full texts reviewed
n=260

Full texts excluded =207
- Conference abstract N=39
- Disease specific mortality N=1
- No disease adjustment N=119
- No relevant intervention/control N=13
- No relevant outcome N=8
- Retracted N=1
- Review or comment N=15
- Second disease N=11

Papers included
n=53
### Figure 2

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<td><strong>0.93 [0.88, 0.99]</strong></td>
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Heterogeneity: Tau² = 0.00, Chi² = 0.30, df = 3 (P = 0.16), I² = 52%

Test for overall effect: Z = 2.21 (P = 0.03)
## Figr-3

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<th>Hazard Ratio IV, Random, 95% CI</th>
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<tbody>
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<td>18.1%</td>
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</tr>
<tr>
<td>Grozaneva 2008</td>
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<td>0.72 [0.65, 0.80]</td>
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<td><strong>Heterogeneity</strong></td>
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<tr>
<td>Tau^2 = 0.00; Chi^2 = 8.16, df = 4 (P = 0.10); P = 35%</td>
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<td>Test for overall effect: Z = 6.40 (P = 0.00001)</td>
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<td><strong>2.1.3 Diet Control</strong></td>
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<td><strong>2.1.4 Insulin Control</strong></td>
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<td>Ekstrom 2012</td>
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<td>Gohda 2013</td>
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<td>Tau^2 = 0.00; Chi^2 = 0.19, df = 1 (P = 0.67); P = 69%</td>
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<td><strong>2.1.5 Sulphonylurea Control</strong></td>
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<td>Kahler 2007</td>
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<td><strong>Heterogeneity</strong></td>
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Test for subgroup differences: Chi^2 = 6.77, df = 3 (P = 0.18); P = 37.2%
### A.

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<td>But 2014</td>
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<td><strong>0.94 [0.92, 0.97]</strong></td>
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Heterogeneity: Tau² = 0.00, Ch² = 0.01, df = 1 (P = 0.92); I² = 0%
Test for overall effect Z = 3.83 (P = 0.0003)

### B.

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Heterogeneity: Tau² = 0.36, Ch² = 22.75, df = 2 (P < 0.0001); I² = 91%
Test for overall effect Z = 8.92 (P < 0.0001)
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<td>0.92 [0.66, 1.00]</td>
</tr>
<tr>
<td>Smiechowski 2013a</td>
<td>-0.17</td>
<td>0.1267</td>
<td>29.0%</td>
<td>0.94 [0.66, 1.08]</td>
</tr>
</tbody>
</table>

Total (95% CI): 100.0% 0.85 [0.72, 1.02]

Heterogeneity: Tau² = 0.01; Chi² = 3.83, df = 2 (P = 0.15); I² = 48%

Test for overall effect: Z = 1.17 (P = 0.28)

### B.

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>log(Hazard Ratio)</th>
<th>SE</th>
<th>Weight</th>
<th>Hazard Ratio IV, Random, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bodimer 2010</td>
<td>-0.5203</td>
<td>0.1989</td>
<td>36.0%</td>
<td>0.59 [0.40, 0.87]</td>
</tr>
<tr>
<td>Bosco 2011</td>
<td>-0.1522</td>
<td>0.1647</td>
<td>47.9%</td>
<td>0.88 [0.62, 1.18]</td>
</tr>
<tr>
<td>Libby 2009</td>
<td>-0.5108</td>
<td>0.3207</td>
<td>18.1%</td>
<td>0.60 [0.32, 1.12]</td>
</tr>
</tbody>
</table>

Total (95% CI): 100.0% 0.71 [0.54, 0.92]

Heterogeneity: Tau² = 0.01; Chi² = 2.49, df = 2 (P = 0.29); I² = 20%

Test for overall effect: Z = 2.55 (P = 0.01)

### C.

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>log(Hazard Ratio)</th>
<th>SE</th>
<th>Weight</th>
<th>Hazard Ratio IV, Random, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Libby 2009</td>
<td>-0.3567</td>
<td>0.2486</td>
<td>13.5%</td>
<td>0.70 [0.43, 1.14]</td>
</tr>
<tr>
<td>Mazzone 2012</td>
<td>-0.3411</td>
<td>0.0994</td>
<td>40.7%</td>
<td>0.71 [0.59, 0.88]</td>
</tr>
<tr>
<td>Smiechowski 2013b</td>
<td>-0.0866</td>
<td>0.0939</td>
<td>45.9%</td>
<td>0.92 [0.78, 1.08]</td>
</tr>
</tbody>
</table>

Total (95% CI): 100.0% 0.80 [0.65, 0.98]

Heterogeneity: Tau² = 0.02; Chi² = 4.26, df = 2 (P = 0.12); I² = 53%

Test for overall effect: Z = 2.21 (P = 0.03)
### Fig 6

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>log(Hazard Ratio)</th>
<th>SE</th>
<th>Weight</th>
<th>IV, Random, 95% CI</th>
<th>Hazard Ratio</th>
<th>IV, Random, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>2.5.1 All Non-Metformin Control</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ekstrom 2012</td>
<td>-0.5965</td>
<td>0.0817</td>
<td>35.7%</td>
<td>0.71 [0.63, 0.81]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gell 2015</td>
<td>-0.434</td>
<td>0.1707</td>
<td>12.8%</td>
<td>0.66 [0.48, 0.90]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ghobati 2013</td>
<td>-0.1744</td>
<td>0.0786</td>
<td>30.8%</td>
<td>0.84 [0.72, 0.98]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Janson 2014</td>
<td>-0.5447</td>
<td>0.2157</td>
<td>5.8%</td>
<td>0.58 [0.38, 0.89]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peters 2013</td>
<td>-0.0101</td>
<td>0.1766</td>
<td>12.1%</td>
<td>0.99 [0.70, 1.40]</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Subtotal (95% CI)</strong></td>
<td>100.0%</td>
<td></td>
<td></td>
<td>0.76 [0.66, 0.87]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Heterogeneity:** Tau² = 0.01, Chi² = 7.23, df = 4 (P = 0.12), I² = 45%

Test for overall effect: Z = 3.63 (P = 0.0001)

<table>
<thead>
<tr>
<th><strong>2.5.2 Diet Control</strong></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Sullivan 2011</td>
<td>0.0677</td>
<td>0.1056</td>
<td>100.0%</td>
<td>1.07 [0.87, 1.32]</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Subtotal (95% CI)</strong></td>
<td>100.0%</td>
<td></td>
<td></td>
<td>1.07 [0.87, 1.32]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Heterogeneity: Not applicable

Test for overall effect: Z = 0.64 (P = 0.52)

<table>
<thead>
<tr>
<th><strong>2.5.3 Insulin Control</strong></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Ekstrom 2012</td>
<td>-0.2463</td>
<td>0.0347</td>
<td>93.0%</td>
<td>0.78 [0.73, 0.84]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ghobati 2013</td>
<td>-0.2993</td>
<td>0.1228</td>
<td>7.0%</td>
<td>0.74 [0.59, 0.95]</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Subtotal (95% CI)</strong></td>
<td>100.0%</td>
<td></td>
<td></td>
<td>0.78 [0.73, 0.84]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Heterogeneity: Tau² = 0.00, Chi² = 0.18, df = 1 (P = 0.69), I² = 0%

Test for overall effect: Z = 7.49 (P = 0.000001)

<table>
<thead>
<tr>
<th><strong>2.5.4 Sulphonylurea Control</strong></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Evans 2009</td>
<td>-0.0813</td>
<td>0.0721</td>
<td>65.7%</td>
<td>0.94 [0.82, 1.08]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sullivan 2011</td>
<td>0.0304</td>
<td>0.0038</td>
<td>34.3%</td>
<td>1.03 [0.85, 1.25]</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Subtotal (95% CI)</strong></td>
<td>100.0%</td>
<td></td>
<td></td>
<td>0.97 [0.87, 1.09]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Heterogeneity: Tau² = 0.00, Chi² = 0.55, df = 1 (P = 0.46), I² = 0%

Test for overall effect: Z = 0.51 (P = 0.61)

Test for subgroup differences: Chi² = 15.29, df = 3 (P = 0.0004), I² = 83.8%
### Fig 7

#### A.

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>log(Hazard Ratio)</th>
<th>SE</th>
<th>Weight</th>
<th>IV, Random, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Floyd 2016</td>
<td>-0.3535</td>
<td>0.1632</td>
<td>84.7%</td>
<td>0.70 [0.52, 0.95]</td>
</tr>
<tr>
<td>Janssone 2014</td>
<td>-0.3425</td>
<td>0.3608</td>
<td>15.3%</td>
<td>0.71 [0.35, 1.44]</td>
</tr>
<tr>
<td>Total (95% CI)</td>
<td></td>
<td></td>
<td>100.0%</td>
<td>0.70 [0.53, 0.93]</td>
</tr>
</tbody>
</table>

Heterogeneity: Tau² = 0.00; Ch² = 0.00, df = 1 (P = 0.99); I² = 0%
Test for overall effect: Z = 2.49 (P = 0.01)

#### B.

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>log(Hazard Ratio)</th>
<th>SE</th>
<th>Weight</th>
<th>IV, Random, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Floyd 2016</td>
<td>-0.8888</td>
<td>0.1214</td>
<td>55.7%</td>
<td>0.92 [0.72, 1.16]</td>
</tr>
<tr>
<td>Janssone 2014</td>
<td>-0.9103</td>
<td>0.305</td>
<td>44.3%</td>
<td>0.90 [0.72, 0.73]</td>
</tr>
<tr>
<td>Total (95% CI)</td>
<td></td>
<td></td>
<td>100.0%</td>
<td>0.63 [0.28, 1.42]</td>
</tr>
</tbody>
</table>

Heterogeneity: Tau² = 0.28; Ch² = 0.36, df = 1 (P = 0.01); I² = 84%
Test for overall effect: Z = 1.11 (P = 0.27)
Table 1. Characteristics of studies investigating the effect of metformin on all cause mortality

<table>
<thead>
<tr>
<th>Study</th>
<th>Exposure</th>
<th>Comparator</th>
<th>Population</th>
<th>N/events and Follow up</th>
<th>Diabes control adjustment</th>
<th>Covariates adjusted for in multivariable analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Metformin compared to Non-Diabetic or General Population</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bannister 2014</td>
<td>Metformin initiation</td>
<td>Non diabetic matched for age, gender, same general practice, prior cancer status and smoking status</td>
<td>United Kingdom. Cohort. Incident diabetes and exposed to glucose lowering therapy for a minimum of 180 days (excluded if any record of secondary diabetes).</td>
<td>N/a</td>
<td>Metformin= 78,241/2,663</td>
<td>3 Non-diabetic= 78,241/2,669 9 deaths Mean: 2.8 years Age, modified Charlson index, gender, smoking status, prior antiplatelet therapy, prior lipid-lowering therapy, prior antihypertensive therapy, year of study index date and study arm.</td>
</tr>
<tr>
<td>Berard 2011</td>
<td>Metformin use at baseline</td>
<td>Non diabetic at baseline</td>
<td>France. Cohort. Middle aged men and women with diabetes. Exposure (hypoglycaemic drug use, presence of metformin) was assessed at entry only.</td>
<td>N/a</td>
<td>Metformin= 40/9</td>
<td>Non-diabetic= 3,162/213 14 years Duration of diabetes, history of diabetes complications, area of residence, age, gender, educational level, alcohol consumption, smoking, blood pressure, LDL and HDL cholesterol.</td>
</tr>
<tr>
<td>Bo 2012</td>
<td>Metformin use</td>
<td>General population based on age and sex</td>
<td>Italy. Cohort. Patients with type 2 diabetes.</td>
<td>N/a</td>
<td>Metformin= NR/104</td>
<td>General population= NR/128 Mean= 4.5 years None.</td>
</tr>
<tr>
<td>Claesen 2016</td>
<td>Metformin use at baseline (censored if discontinued for more than 9 months)</td>
<td>Non users of glucose lowering agents</td>
<td>Belgium. Cohort. Patients older than 18 prescribed metformin and non users of glucose lowering agents matched 5 to one on age, gender, cardiovascular history, associated therapies and year of start of follow up.</td>
<td>N/a</td>
<td>Metformin= 42,900/3,389</td>
<td>9 Non-diabetic= 214,500/16,517 5 years Adjusted for age, associated therapies, and gender.</td>
</tr>
<tr>
<td><strong>Metformin compared to other diabetes therapies, controlling for diabetes management</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bo 2012</td>
<td>Metformin use</td>
<td>Management of diabetes through diet alone</td>
<td>Italy. Cohort. Patients with type 2 diabetes.</td>
<td>HbA1c</td>
<td>Metformin= 1,479/136 Diet= 620/68 Mean: 4.5 years Adjusted for propensity score which included: propensity score which included: age, sex, diabetes duration, HbA1c, smoking, BMI, retinopathy, nephropathy, coronary or peripheral artery disease, other comorbidities, use of hypertensives, use of acetyl salicylic acid</td>
<td></td>
</tr>
<tr>
<td>Ekstrom 2012</td>
<td>Metformin use continuously for 12 months</td>
<td>Insulin use continuously for 12 months</td>
<td>Sweden. Cohort. Men and women aged 40 to 85 with type 2 diabetes on</td>
<td>HbA1c at baseline (at least 1 year after)</td>
<td>Metformin= 14,697/1734</td>
<td>Age, sex, diabetes duration, HbA1c, non-HDL-C, BMI, smoking, eGFR, multidose</td>
</tr>
<tr>
<td>Study</td>
<td>Intervention</td>
<td>Matched Controls</td>
<td>Study Design</td>
<td>Eligibility Criteria</td>
<td>Average Change in HbA1c over 24 months</td>
<td>Covariates Adjusted for</td>
</tr>
<tr>
<td>-------</td>
<td>--------------</td>
<td>------------------</td>
<td>--------------</td>
<td>----------------------</td>
<td>----------------------------------------</td>
<td>------------------------</td>
</tr>
<tr>
<td>Evans 2006</td>
<td>Metformin monotherapy throughout study period (initiated at baseline, censored from date of switch to new therapy)</td>
<td>Sulfonylurea monotherapy through study period (initiated at baseline, censored from date of switch to new therapy)</td>
<td>United Kingdom. Cohort. Patients with type 2 diabetes newly prescribed OHAs.</td>
<td>Age, gender, Diabetes duration, previous hospitalisation, history of CVD and CHF, microalbuminuria, and treatment with antihypertensive agents, lipid lowering agents, and cardiac glycosides</td>
<td>Metformin=2,286/107 Sulphonylur ea=3,331/597 NR</td>
<td>Gender, Age, Diabetes duration, previous CV hospitalisation, Smoking, mean HbA1c, mean BMI, mean systolic blood pressure, mean diastolic blood pressure, mean cholesterol, using aspirin, using statins, using beta blockers, using ACE inhibitors.</td>
</tr>
<tr>
<td>Ghotbi 2013</td>
<td>Metformin monotherapy or Metformin with insulin</td>
<td>Insulin monotherapy OR Any other therapy (Diet, insulin, sulphonylurea) alone or in any combination</td>
<td>Worldwide (16 countries). Cohort. Patients who were 55 years or over with BMI 27-45 with diabetes.</td>
<td>Age, smoking habits, diabetes duration, congestive heart failure, history of hypertension, BMI, sex, history of CVD, tobacco use, HDL concentrations, LDL concentrations, change in HbA1c values and/or HbA1c levels, heart rate, actual systolic and diastolic blood pressure, sibutramine usage</td>
<td>Metformin=1,631/96 Metformin with insulin=1000/88 Insulin=1,116/138 Any other therapy= NR</td>
<td>Age, smoking habits, diabetes duration, congestive heart failure, history of hypertension, BMI, sex, history of CVD, tobacco use, HDL concentrations, LDL concentrations, change in HbA1c values and/or HbA1c levels, heart rate, actual systolic and diastolic blood pressure, sibutramine usage</td>
</tr>
<tr>
<td>Gosmanova 2008</td>
<td>Metformin (combination or monotherapy)</td>
<td>Not receiving metformin</td>
<td>USA. Cohort. Veterans with type 2 diabetes receiving long term care at veterans affair medical center treated with a prescription claim for oral hypoglyemic agents</td>
<td>Age, race, eGFR, HbA1c, use of insulin, ACEI/ARBs, or statins.</td>
<td>Metformin=1,207/266, Non-metformin=999/253 Metformin=62 months, Non-metformin=61 months</td>
<td>Age, race, eGFR, HbA1c, use of insulin, ACEI/ARBs, or statins.</td>
</tr>
<tr>
<td>Kahler 2007</td>
<td>Metformin monotherapy</td>
<td>Sulfonylurea monotherapy</td>
<td>USA. Cohort. People with diabetes at least 18 years of age who survived at least 1 year after drug assessment.</td>
<td>Age, diabetes suration, HbA1C, creatinine, diabetes related physician visits and also a propensity score constructed from 48 variables.</td>
<td>Metformin=2,988/82 Metformin with sulphonylur ea=13,820/468 Sulphonylur ea=19,053/1,00 NR</td>
<td>Age, diabetes suration, HbA1C, creatinine, diabetes related physician visits and also a propensity score constructed from 48 variables.</td>
</tr>
<tr>
<td>Author</td>
<td>Year</td>
<td>Study Design</td>
<td>Country</td>
<td>Diagnosis Criteria</td>
<td>HbA1C during the Study Period</td>
<td>Metformin Use</td>
</tr>
<tr>
<td>--------</td>
<td>------</td>
<td>--------------</td>
<td>---------</td>
<td>--------------------</td>
<td>-------------------------------</td>
<td>--------------</td>
</tr>
<tr>
<td>Libby</td>
<td>2009</td>
<td>In receipt of a metformin prescription</td>
<td>United Kingdom. Cohort. Patients diagnosed with type 2 diabetes aged 35 or over.</td>
<td>HbA1C during the study period</td>
<td>Metformin=1,085/609 Non-metformin=4,085/1,422</td>
<td>Age, sex, smoking, deprivation, BMI, A1C, insulin use, sulphonylurea use.</td>
</tr>
<tr>
<td>Pratipan awatr</td>
<td>2010</td>
<td>Metformin use</td>
<td>Thailand. Cohort. Diabetic patients receiving clinic treatment</td>
<td>HbA1C at baseline (treatment was ongoing)</td>
<td>Overall=9,370/424 3 years</td>
<td>Age, sex, HbA1c, serum creatinine, healthcare plan, education status, smoking status, previous history of coronary artery disease and cerebrovascular disease, lipid lowering medication, insulin.</td>
</tr>
<tr>
<td>Sullivan</td>
<td>2011</td>
<td>Metformin monotherapy</td>
<td>International. Cohort. Patients with type 2 diabetes</td>
<td>HbA1c at baseline (treatment was ongoing)</td>
<td>Metformin=1,746/NR Diet=1,632 Sulphonylurea=1,632 5 years</td>
<td>Age, sex, duration of diabetes, smoking, waist-hip ratio, systolic blood pressure, total cholesterol, HDL cholesterol, HbA1c, ACR group, history of CVD, presence of microvascular disease, creatinine and peripheral damage to feet.</td>
</tr>
<tr>
<td>Wang</td>
<td>2014</td>
<td>Metformin prescription as sole class of glucose lowering medication for ≥180 days</td>
<td>USA. Cohort. Veterans aged 65-89 years with type 2 diabetes without history of liver, renal diseases, or cancers</td>
<td>HbA1c during the study period</td>
<td>Metformin=307/NR Sulphonylurea=2,108 Mean=5.6 years</td>
<td>Age, race, diabetes duration, age adjusted Charlson co-morbidity, smoking cessation status, mean LDL across study period, mean HbA1c across study period and propensity score.</td>
</tr>
<tr>
<td>Study</td>
<td>Exposure</td>
<td>Comparator</td>
<td>Population</td>
<td>Diabetes control adjustment</td>
<td>N/events and Follow up</td>
<td>Covariates adjusted for in multivariable analysis</td>
</tr>
<tr>
<td>---------------</td>
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<td>-----------------------------------------</td>
<td>-------------------------------------------------</td>
<td>-----------------------------</td>
<td>------------------------</td>
<td>--------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Andersson 2012</td>
<td>Metformin compared to Non-Diabetic or General Population</td>
<td>Non users of glucose lowering agents (presumed non-diabetic).</td>
<td>Denmark. Cohort. Individuals who had never had cancer aged ≥35 years</td>
<td>N/a</td>
<td>Cancer incidence Metformin=11,359/5,450 Non-diabetic=3,447,904/293,878</td>
<td>Charlson comorbidity score, gender, age, calendar year</td>
</tr>
<tr>
<td>But 2014</td>
<td>Metformin use</td>
<td>Non-users of anti-diabetic medications (presumed non-diabetic).</td>
<td>Finland. Cohort. Stratified randomly drawn sample (sex, 10 year age groups, geographical area).</td>
<td>N/a</td>
<td>Cancer incidence Metformin=1,88/42 Non-diabetic=22,093/1,029</td>
<td>Age, gender, calendar time, BMI, smoking status, interaction of age and gender, age and BMI. Median 9 years</td>
</tr>
<tr>
<td>Tseng 2011</td>
<td>Metformin use</td>
<td>General population</td>
<td>Taiwan. Cohort. People without prior bladder cancer</td>
<td>N/a</td>
<td>Bladder cancer Overall=1,000,000/589 2 years</td>
<td>Age, sex, diabetes, nephropathy, urinary tract disease, Sulfonylurea, Acarbose, THZ, Insulin, hypertension, COPD, Stroke, ischemic heart disease, peripheral arterial disease, eye disease, dyslipidaemia, statin, fibrate, ACE inhibitor, calcium channel blocker, living region, occupation</td>
</tr>
<tr>
<td>Tseng 2012a</td>
<td>Metformin use</td>
<td>General population</td>
<td>Taiwan. Cohort. People without type 1 diabetes</td>
<td>N/a</td>
<td>Thyroid cancer Overall=99, 739 Metformin=NR/37 Gen Pop=NR/906</td>
<td>Age, sex, comorbidities, medications, living region, occupation, potential detection examinations</td>
</tr>
<tr>
<td>Tseng 2012b</td>
<td>Metformin use</td>
<td>General population</td>
<td>Taiwan. Cohort. People without type 1 diabetes</td>
<td>N/a</td>
<td>Colorectal cancer Overall=995,843</td>
<td>Age, sex, diabetes status, dyslipidaemia, obesity, hypertension, COPD, asthma, stroke,</td>
</tr>
<tr>
<td>Study</td>
<td>Exposure Duration</td>
<td>Metformin Use</td>
<td>Never Use of Metformin</td>
<td>United Kingdom</td>
<td>Outcome</td>
<td>Other Factors</td>
</tr>
<tr>
<td>--------------</td>
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<td>---------------</td>
<td>------------------------</td>
<td>----------------</td>
<td>--------------------------------</td>
<td>-------------------------------------------------</td>
</tr>
<tr>
<td>Becker 2014</td>
<td>Any metformin use</td>
<td>N/a</td>
<td>N/a</td>
<td>N/a</td>
<td>Head and neck cancer</td>
<td>All other medications (sulphonylurea, insulin and TZD), BMI, smoking, and diagnosis of diabetes melitus</td>
</tr>
<tr>
<td>Short term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Long term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Becker 2016</td>
<td>Any metformin use</td>
<td>N/a</td>
<td>N/a</td>
<td>N/a</td>
<td>Renal cell carcinoma</td>
<td>All other medications (sulphonylurea, insulin and TZD), BMI, smoking, alcohol consumption, hypertension and diagnosis of diabetes melitus</td>
</tr>
<tr>
<td>Short term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Long term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Becker 2013</td>
<td>Metformin exposure</td>
<td>N/a</td>
<td>N/a</td>
<td>N/a</td>
<td>Esophageal cancer</td>
<td>Other medications (Sulphonylureas, Insulin, TZD), BMI, and smoking.</td>
</tr>
<tr>
<td>Short term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Medium term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Long term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bodmer 2012a</td>
<td>Metformin exposure</td>
<td>N/a</td>
<td>N/a</td>
<td>N/a</td>
<td>Lung cancer</td>
<td>Other drug use (Sulphonylureas, insulin), BMI and smoking.</td>
</tr>
<tr>
<td>Short term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Medium term</td>
<td>N/a</td>
<td></td>
<td>N/a</td>
<td>N/a</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Notes:**
- Metformin exposure:
  - Short term (1-14 prescription):
  - OR
  - Medium term (15-29 prescription):
  - OR
  - Long term (≥30 prescription):
  - OR
- Never use of metformin:
  - United Kingdom:
  - Case-control.
  - Patients aged 40 to 89 excluding those with HIV, alcoholism, or any malignancy prior to the index date. Controls matched for calendar time, age, sex, general practice, number of years active history with up to 10 controls.
- Outcome:
  - Head and neck cancer
  - Renal cell carcinoma
  - Esophageal cancer
- Other factors:
  - All other medications (sulphonylurea, insulin and TZD), BMI, smoking, alcohol consumption, hypertension and diagnosis of diabetes melitus
  - Other medications (Sulphonylureas, Insulin, TZD), BMI, and smoking.
<table>
<thead>
<tr>
<th>Study</th>
<th>Exposure Details</th>
<th>Control Details</th>
<th>Duration Details</th>
<th>Cancer Prevalence</th>
<th>Adjusted For</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bodmer 2012b</td>
<td>Metformin exposure</td>
<td>No use of metformin</td>
<td>United Kingdom. Case-control. Participants were below the age of 90 years had no history of other cancers, alcoholism or HIV. Controls matched 6:1 on age, sex, calendar time, general practice, and years of history. Adjusted for other medications (insulin of sulphonylurea), BMI, smoking, alcohol consumption and diabetes duration.</td>
<td>Pancreatic cancer</td>
<td>Age, sex, body mass index, smoking, alcohol drinking, Townsend deprivation index, calendar year, GP visit 1 year before index date, and diabetes (when appropriate for antidiabetics).</td>
</tr>
<tr>
<td>Lu 2015</td>
<td>Use of metformin</td>
<td>Non diabetic</td>
<td>United Kingdom. Case-control. Patients without a history of cancer or prior pancreatic disease aged 20 to 79 years. Controls frequency matched for sex, age, and calendar year.</td>
<td>Pancreatic cancer</td>
<td>Age, sex, race, BMI, history of pancreatitis, alcohol, smoking, family history of PC, and diabetes medications.</td>
</tr>
<tr>
<td>Walker 2012</td>
<td>Ever use of metformin</td>
<td>Never use of metformin (non-diabetic)</td>
<td>USA. Case control. Patients aged 21 to 85 years. Patients with diabetes diagnosed within 1 year of index excluded. Controls were frequency matched by sex, and age.</td>
<td>Pancreatic cancer</td>
<td>Age, sex, race, BMI, history of pancreatitis, alcohol, smoking, family history of PC, and diabetes medications.</td>
</tr>
<tr>
<td>Baur 2011</td>
<td>Metformin monotherapy</td>
<td>Non type 2 diabetic</td>
<td>Germany. Cross-sectional. Consecutive primary care patients HbA1c (and non-diabetic comparators)</td>
<td>Cancer prevalence</td>
<td>Adjusted for age, sex, HbA1c, smoking status and BMI</td>
</tr>
<tr>
<td>Nordstrom 2015</td>
<td>Metformin (any within 2 years of biopsy)</td>
<td>Non users of any antidiabetic medication (presumed non-diabetic)</td>
<td>Sweden. Cross-sectional. Men undergoing their first prostate biopsy</td>
<td>Prostate cancer</td>
<td>Adjusted for age, natural log transformed PSA concentration, PSA quotient, Charlson comorbidity index,</td>
</tr>
</tbody>
</table>
### Metformin compared to other diabetes therapies, controlling for diabetes management

<table>
<thead>
<tr>
<th>Study</th>
<th>Design</th>
<th>Exposure</th>
<th>Comparator</th>
<th>Outcome</th>
<th>Matched Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gossens 2015</td>
<td>In receipt of a metformin prescription</td>
<td>Metformin use (measured throughout the study period)</td>
<td>Control</td>
<td>Bladder cancer</td>
<td>Age, gender, smoking, BMI, HbA1c across study period</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Only metformin use</td>
<td>Cancer incidence, colorectal cancer, lung cancer, breast cancer</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Metformin with sulphonylurea</td>
<td>Metformin=4,085/297, 40, 35, or 24 Non-metformin=4,085/474, 76, 58, or 41</td>
<td></td>
</tr>
<tr>
<td>Libby 2009</td>
<td>Continuous prescription for at least 6 months of metformin monotherapy</td>
<td>Matched patients (year of diagnosis) who had no record of metformin use</td>
<td>Control</td>
<td>Prostate cancer</td>
<td>Age, period, region, BMI, year of diagnosis and weighted HbA1c</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Metformin with sulphonylurea</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Continuous prescription for at least 6 months of sulphonylurea monotherapy</td>
<td></td>
<td>Prostate cancer</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>United Kingdom. Cohort. Women with incident diabetes without a previous diagnosis of breast cancer</td>
<td></td>
<td>Prostate cancer</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Time weighted average of HbA1c</td>
<td></td>
<td>Prostate cancer</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Breast cancer</td>
<td></td>
<td>Prostate cancer</td>
<td></td>
</tr>
</tbody>
</table>

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**Gossens 2015**
- Metformin use (measured throughout the study period)
- Control or metformin use
- Bladder cancer
- Matched patients (year of diagnosis) who had no record of metformin use
- Cancer incidence, colorectal cancer, lung cancer, breast cancer
- Age, gender, smoking, BMI, HbA1c across study period

**Libby 2009**
- In receipt of a metformin prescription
- Matched patients (year of diagnosis) who had no record of metformin use
- Prostate cancer
- Time weighted average of HbA1c
- Breast cancer
- Age, period, region, BMI, year of diagnosis and weighted HbA1c

**Redaniel 2012**
- Continuous prescription for at least 6 months of metformin monotherapy
- Continuous prescription for at least 6 months of sulphonylurea monotherapy
- United Kingdom. Cohort. Women with incident diabetes without a previous diagnosis of breast cancer
- Time weighted average of HbA1c
- Breast cancer
- Age, period, region, BMI, year of diagnosis and weighted HbA1c

**Azoulay 2011**
- Ever use of metformin (at least on prescription prior to index)
- Ever use of any other antidiabetic agent.
- United Kingdom. Case-control. Male patients who with incident use of at least one antidiabetic agent (not insulin) aged 40 years or greater. Controls were matched at up to 10:1 after matching for year of birth, and date of cohort entry.
- HbA1c (last measurement before index date)
- Prostate cancer
- HbA1c at index, excessive alcohol use, obesity, smoking, lower urinary tract symptoms, previous cancer, previous use of NSAIDs, antihypertensives or statins, and ever use of other antidiabetics
<table>
<thead>
<tr>
<th>Becker 2013</th>
<th>Metformin exposure</th>
<th>No use of metformin</th>
<th>United Kingdom. Case-control. Patients aged 40 to 89 with diabetes excluding those with HIV, alcoholism, or any malignancy prior to the index date. Controls matched up to 10:1 for calendar time, age, sex, general practice, number of years active history.</th>
<th>HbA1c</th>
<th>Esophageal cancer</th>
<th>Other medications (Sulphonylurea, Insulin, TZD), BMI, smoking, diabetes duration and HbA1c level.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Short term (1-14 prescription s) OR Medium term (15-29 prescription s) OR Long term (≥30 prescription s)</td>
<td>No use of metformin</td>
<td>United Kingdom. Case-control. Patients aged 40 to 89 with diabetes excluding those with HIV, alcoholism, or any malignancy prior to the index date. Controls matched up to 10:1 for calendar time, age, sex, general practice, number of years active history.</td>
<td>HbA1c</td>
<td>Esophageal cancer</td>
<td>Other medications (Sulphonylurea, Insulin, TZD), BMI, smoking, diabetes duration and HbA1c level.</td>
<td></td>
</tr>
<tr>
<td>Bodmer 2010</td>
<td>Metformin use Short term (1-9 prescription s) Medium term (10-39 prescription s) Long term (≥40 prescription s)</td>
<td>No use of metformin</td>
<td>United Kingdom. Case-control. Patients aged 40 to 89 with diabetes excluding those with HIV, alcoholism, or any malignancy prior to the index date. Controls matched up to 10:1 for calendar time, age, sex, general practice, number of years active history.</td>
<td>Last reported HbA1C</td>
<td>Breast cancer</td>
<td>Sulphonylurea, THZ, Insulin, prandial glucose regulators, acarbose, estrogens, smoking, BMI, diabetes duration, and HbA1C.</td>
</tr>
<tr>
<td>Bosco 2011</td>
<td>Metformin use (redeemed prescription s for at least 1 year) Or Metformin use (redeemed prescription s for at least 5 year2)</td>
<td>No metformin Or Other anti diabetic medication OR Diet/exercise only</td>
<td>Denmark. Case-control. Women with type 2 diabetes aged 50 or over. Controls matched 10:1 by risk set sampling from the same county</td>
<td>Diabetes complications</td>
<td>Breast cancer ≥1 year metformin= 1250/96 ≥ 5 year metformin= 453/35 No metformin= 297/2803 Other medications = 2197/219 Diet/exercise= 876/78</td>
<td>County, complications due to diabetes, clinical obesity, age at index date, postmenopausal hormone use, and multiple imputation to impute missing parity.</td>
</tr>
<tr>
<td>Chen 2013</td>
<td>Any use of metformin</td>
<td>No use of metformin</td>
<td>Taiwan. Case-control. People with diabetes excluding those with previous malignancies, aged younger than 20, or who had undergone previous liver surgery. Controls matched 10:1 by risk set sampling from the same county</td>
<td>Physician visits per year</td>
<td>Hepatocellular carcinoma Cases with diabetes= 22,047, Controls with diabetes= 25,773</td>
<td>Age, gender, hepatitis B, hepatitis C, liver cirrhosis, end stage renal disease, DM duration, DM control (assessed by physician visits), use of other diabetic agents.</td>
</tr>
<tr>
<td>Mazzone 2012</td>
<td>Use of metformin</td>
<td>Non use of metformin</td>
<td>USA. Case-control. Patients with diabetes mellitus.</td>
<td>Mean HbA1c</td>
<td>Lung cancer Overall 1014</td>
<td>BMI, HbA1c, pack years of smoking.</td>
</tr>
<tr>
<td>Study</td>
<td>Follow-up Details</td>
<td>Outcomes</td>
<td>Most Recent HbA1c</td>
<td>Most Recent HbA1c Details</td>
<td></td>
<td></td>
</tr>
<tr>
<td>-------</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Sehdev 2015</td>
<td>Metformin in the 12 months before index date</td>
<td>USA. Case-control. Patients aged &gt;18 years with diabetes mellitus. Controls matched for age, sex, and geographical region</td>
<td>Colorectal cancer</td>
<td>Metformin=3,042/983 Non use=5,004/1,699</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Number of hospital admissions and number of outpatient visits.</td>
<td>12 months</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Obesity, polycystic ovary disease, inflammatory bowel disease, sulphonylurea use, coronary artery disease, prescribed NSAIDs, insulin use, TZD use, Charlson comorbidity index, number of hospital admissions, number of outpatient visits.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smiechowski 2013a</td>
<td>Ever use of metformin (at least 12 months prior to index)</td>
<td>United Kingdom. Case-control. Patients at least 40 years of age who had received at least one non-insulin antidiabetic prescription. Controls were matched 10:1 for age, sex, calendar year of cohort entry, and duration of follow up.</td>
<td>Colorectal cancer</td>
<td>Metformin=1,594/163 Non use=5,450/44</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Most recent HbA1c</td>
<td>Mean 4.8 years</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Obesity, smoking, statins, NSAIDs, aspirin, excessive alcohol use, HbA1c, diabetes duration, cholecystectomy, inflammatory bowel disease, referrals to colonoscopy, referrals to sigmoidoscopy, history of polyps, previous cancer, use of sulphonylurea, THZs, insulins and other antidiabetic agents.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smiechowski 2013b</td>
<td>Ever use of metformin (at least 12 months prior to index)</td>
<td>United Kingdom. Case-control. Patients at least 40 years of age who had received at least one non-insulin antidiabetic prescription. Controls matched 10:1 for age, sex, calendar year, and duration of follow up.</td>
<td>Lung cancer</td>
<td>Metformin=6,611/616 Non use=1,961/192</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Most recent HbA1c</td>
<td>Mean=5.0 years</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Diabetes duration, HbA1c, obesity, smoking, excessive alcohol use, previous cancer, COPD, asthma, NSAIDs, aspirin, statins, other antidiabetic drugs.</td>
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</tbody>
</table>
Supp table 2. Characteristics of studies investigating the effect of metformin on all cardiovascular disease

<table>
<thead>
<tr>
<th>Study</th>
<th>Exposure</th>
<th>Comparator</th>
<th>Population</th>
<th>Diabetes control adjustment</th>
<th>N/events and Follow up</th>
<th>Covariates adjusted for in multivariable analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ekestrom 2012</td>
<td>Metformin compared to other diabetes therapies, controlling for diabetes management</td>
<td>Insulin use continuously for 12 months before baseline or Other OHA or Insulin and other OHA</td>
<td>Sweden. Cohort. Men and women aged 40 to 85 with type 2 diabetes on continuous glucose lowering treatment that filed at least 3 prescriptions for their treatment (or 18 fills of multi-dose dispensed drugs) in the 12 months before baseline.</td>
<td>HbA1c at baseline (at least 1 year after treatment commence ment)</td>
<td>CVD (Any) Metformin= 14,317/1,734 Insulin=11,427/2,389 Other OHA=4,964/929 Insulin + Other OHA=1,365/NR</td>
<td>Age, sex, diabetes duration, HbA1c, non-HDL-C, BMI, smoking, eGFR, multidose dispensation, previous hospitalisation, history of CVD and CHF, microalbuminuria, and treatment with antihypertensive agents, lipid lowering agents, and cardiac glycosides</td>
</tr>
<tr>
<td>Evans 2006</td>
<td>Metformin monotherapy throughout study period (initiated at baseline, censored from date of switch to new therapy)</td>
<td>Sulfonylurea monotherapy throughout study period (initiated at baseline, censored from date of switch to new therapy)</td>
<td>United Kingdom. Cohort. Patients with type 2 diabetes newly prescribed OHA.</td>
<td>Average HbA1c for the study period</td>
<td>CVD admission (Any) Metformin= 2,286/229 Sulfonylurea= 3,331/567 NR</td>
<td>Gender, Age, Diabetes duration, previous CV hospitalisation, Smoking, mean HbA1c, mean BMI, mean systolic blood pressure, mean diastolic blood pressure, mean cholesterol, using aspirin, using statins, using beta blockers, using ACE inhibitors.</td>
</tr>
<tr>
<td>Jansson 2014</td>
<td>Treatment with metformin</td>
<td>Non users of metformin</td>
<td>Sweden. Cohort. Patients with incident diabetes during the study period</td>
<td>Blood glucose level (annual mean values)</td>
<td>CVD (Any) Metformin= 189/NR Non-metformin= 951/NR Myocardial infarction Overall events= 298 Stroke overall events = 565</td>
<td>Age, sex, year of diabetes diagnosis, BMI, smoking habits, blood pressure, blood glucose level, number of previous events of the outcome under study, diabetes treatment, blood pressure treatment.</td>
</tr>
<tr>
<td>McAllister 2008</td>
<td>Dispensation of metformin exclusively through the duration of the study</td>
<td>Dispensation of sulphonylurea exclusively through the duration of the study</td>
<td>Canada. Cohort. Aged 30 years or over with at least 1 year of continuous coverage who did not have a diagnosis of heart failure at baseline.</td>
<td>Total physician visits after diabetic medication index dispensation</td>
<td>Heart failure Metformin monotherapy γ= 1,469/208 Sulphonylurea monotherapy γ= 4,162/77</td>
<td>Age, gender, modified chronic disease score, therapies known to affect heart failure occurrence or strongly correlated with factors predicting heart failure, and total physician visits after diabetic medication index dispensation.</td>
</tr>
<tr>
<td>Study</td>
<td>Type</td>
<td>Cohort</td>
<td>Population</td>
<td>HbA1c (at event or censoring)</td>
<td>Conditions</td>
<td>Follow-up</td>
</tr>
<tr>
<td>-------</td>
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<td>------------</td>
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</tr>
<tr>
<td>Nichols 2005</td>
<td>Metformin monotherapy</td>
<td>USA. Cohort. People with a diagnosis of diabetes that did not have a history of congestive heart failure.</td>
<td>HbA1c (at event or censoring)</td>
<td>Congestive heart failure</td>
<td>Mean 4.6-4.7 years</td>
<td>Age, sex, suration of diabetes, HbA1c, ischemic heart disease, hypertension, renal insufficiency</td>
</tr>
<tr>
<td>Peters 2013</td>
<td>Use of metformin</td>
<td>Australia. Cohort. Patients with type 2 diabetes who were not using insulin at baseline</td>
<td>Baseline HbA1c (drug use was ongoing)</td>
<td>Coronary heart disease</td>
<td>Mean 23-26 months</td>
<td>There was conditional variable selection but considered variables were age, sex, diabetes duration, BMI, waist circumference, fasting serum glucose, HbA1c, Serum magnesium, hypomagnesium, urinary magnesium, fractional excretion urinary magnesium, renal causes, serum creatinine, Alcohol consumption, diuretic therapy, digoxin therapy, magnesium supplementation, gastrointestinal problems.</td>
</tr>
<tr>
<td>Sullivan 2011</td>
<td>Metformin monotherapy</td>
<td>International. Cohort. Patients with type 2 diabetes</td>
<td>HbA1c at baseline (treatment was ongoing)</td>
<td>Coronary heart disease</td>
<td>Mean 12.3 years</td>
<td>Age, sex, duration of diabetes, smoking, waist-hipratio, systolic blood pressure, total cholesterol, HDL cholesterol, HbA1c, ACR group, history of CVD, presence of microvascular disease, creatinine and peripheral damage to feet.</td>
</tr>
<tr>
<td>Ghotbi 2013</td>
<td>Metformin monotherapy Or Metformin + Insulin Or Metformin alone or in combination with any other therapy</td>
<td>Worldwide. Cohort. Patients who were 55 years or over with BMI 27-45 with diabetes.</td>
<td>Change in HbA1c values and/or HbA1c levels</td>
<td>CVD (Nonfatal myocardial infarction, nonfatal stroke, resuscitation after cardiac arrest, and cardiovascul ar death)</td>
<td>Mean 5 years</td>
<td>Age, smoking habits, diabetes duration, congestive heart failure, history of hypertension, BMI, sex, history of CVD, tobacco use, HDL concentrations, LDL concentrations, Change in HbA1c values and/or HbA1c levels, heart rate, actual systolic and diastolic blood pressure, sibutramine usage</td>
</tr>
<tr>
<td>Study</td>
<td>Use of Metformin</td>
<td>Non Use of Metformin</td>
<td>USA / Denmark</td>
<td>Hba1c</td>
<td>CVD</td>
<td>Adjusted for:</td>
</tr>
<tr>
<td>----------</td>
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</tr>
<tr>
<td>Floyd</td>
<td>Current use</td>
<td>Never use</td>
<td>Case-control. People with diabetes aged 30-79 who were users of long lasting insulin. Controls were frequency matched on age, sex and calendar year</td>
<td>HbA1C</td>
<td>Myocardial infarction</td>
<td>Hba1C, index year, age, sex, hypertension status, smoking, prior cardiovascular disease, atrial fibrillation, nephrotic syndrome, diastolic blood pressure, body mass index, total cholesterol, serum creatinine, duration of diabetes, and the use of statins, ACE inhibitors, and regular and rapid-acting insulin</td>
</tr>
<tr>
<td>Gejl</td>
<td>Use of</td>
<td>Non use</td>
<td>Case-control. Patients with diabetes mellitus. Controls were matched 3:1 on gender</td>
<td>HbA1c</td>
<td>CVD (ischemic heart disease, heart failure or stroke)</td>
<td>Adjusted for: previous CE, age, diabetes duration, gender, atrial fibrillation, hypertension, alcohol related diagnosis, nephropathy, retinopathy, neuropathy, peripheral artery disease, usage of antiarrhythmic drugs, vitamin K antagonists, heparin, pentasaccharide, argatroban, thrombocyte function inhibitors, acetylsalicylic acid, cyclooxygenase 2 inhibitors, nonselective cyclooxygenase inhibitors, buprenorphine, tramadol, oxycodone, morphine, codeine, fentanyl, pethidine, glucocorticoids, bisphosphonates, benzodiazepines, antipsychotics, antiepileptic drugs, statins, antidepressants, insulins, glitazones, DPP-4 inhibitors, liraglutide, exenatide, biguanide, and β cell stimulants, as well as: LDL, HDL, total cholesterol, HbA1C, creatinine and triglycerides.</td>
</tr>
<tr>
<td>Hartung</td>
<td>Use of</td>
<td>Use of another OHA</td>
<td>Case-control. Patients who had been hospitalized aged 18 years or older with a diagnosis of diabetes mellitus. Controls were frequency matched 6:1 for age and sex.</td>
<td>Number of previous diabetes-related office visits</td>
<td>Heart failure (hospitalisations)</td>
<td>Charlson comorbidity score and number of previous diabetes related office visits</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Study</th>
<th>Use of</th>
<th>Use of another OHA</th>
<th>USA. Case-control. People who had been hospitalized aged 18 years or older with a diagnosis of diabetes mellitus. Controls were frequency matched 6:1 for age and sex.</th>
<th>Number of previous diabetes-related office visits</th>
<th>Number of previous diabetes-related office visits</th>
<th>Charlson comorbidity score and number of previous diabetes related office visits</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Metformin</td>
<td>Other OHA</td>
<td>USA. Case-control. People who had been hospitalized aged 18 years or older with a diagnosis of diabetes mellitus. Controls were frequency matched 6:1 for age and sex.</td>
<td>Number of previous diabetes-related office visits</td>
<td>Heart failure (hospitalisations)</td>
<td>Charlson comorbidity score and number of previous diabetes related office visits</td>
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<td>USA. Case-control. People who had been hospitalized aged 18 years or older with a diagnosis of diabetes mellitus. Controls were frequency matched 6:1 for age and sex.</td>
<td>Number of previous diabetes-related office visits</td>
<td>Heart failure (hospitalisations)</td>
<td>Charlson comorbidity score and number of previous diabetes related office visits</td>
</tr>
<tr>
<td>Study</td>
<td>Intervention</td>
<td>Comparator</td>
<td>Setting</td>
<td>Methodology</td>
<td>Comparator Event Rate</td>
<td>Comparator Event Rate</td>
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<tr>
<td>Koro 2005</td>
<td>Prescription for metformin</td>
<td>No drug exposure</td>
<td>United Kingdom. Case-control.</td>
<td>Matched case-control</td>
<td>1,067/ 152</td>
<td>2,401/296</td>
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<tr>
<td></td>
<td>Prescription for sulphonylurea</td>
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<td>6:1 for age, gender, calendar year</td>
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<td>and length of history.</td>
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<td>Adjusted for micro and macro</td>
<td>4,138/591</td>
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<td>vascular complications of diabetes</td>
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<td>(retinopathy, neuropathy,</td>
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<td>nephropathy, foot ulcers,</td>
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<td>gangrene and end stage renal</td>
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<td>disease).</td>
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<td>Congestive heart failure</td>
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<td>Metformin= 1,067/ 152</td>
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<td>No drug exposure= 2,401/296</td>
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<td>Sulphonylurea= 4,138/591</td>
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<td>Mean= 3.4</td>
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<td>Gender, index year of CHF</td>
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<td>diagnosis, duration of diabetes,</td>
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<td>infarction, ischemic heart</td>
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<td>disease, peripheral vascular</td>
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<td>disease, valvular disease,</td>
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<td>hypertension, retinopathy,</td>
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<td>nephropathy, foot ulcers,</td>
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<td>gangrene, end stage renal</td>
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<td></td>
<td>disease).</td>
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<tr>
<td>Kooy 2009</td>
<td>Metformin (850mg, 1-3 times</td>
<td>Placebo, with insulin</td>
<td>The Netherlands. RCT. Patients with</td>
<td>Changes in HbA1c</td>
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<tr>
<td></td>
<td>daily) with insulin therapy</td>
<td>therapy</td>
<td>type 2 diabetes mellitus, aged 30-80</td>
<td>Macrovascular morbidity or</td>
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<td>mortality</td>
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<td>Metformin= 196/ NR</td>
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<td>Placebo= 194/ NR</td>
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<td></td>
<td></td>
<td></td>
<td>4.3 years</td>
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<tr>
<td>Giorda 2011</td>
<td>Metformin use</td>
<td>No metformin use</td>
<td>Italy. Cross-sectional. Non-</td>
<td>Changes in HbA1c level, daily</td>
<td>Age, waist circumference,</td>
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<td></td>
<td></td>
<td></td>
<td>institutionalised persons aged &gt;45</td>
<td>dose of insulin, and systolic BP,</td>
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<td>years with diabetes mellitus free</td>
<td>in addition to age, sex, smoking</td>
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<td>from symptoms and clinical signs of</td>
<td>and cardiovascular history</td>
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<td></td>
<td></td>
<td>cardiac disease</td>
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<tr>
<td>Solini 2013</td>
<td>Metformin alone or in</td>
<td>No use of metformin</td>
<td>Italy. Cross-sectional. Individuals</td>
<td>HbA1c</td>
<td>Age, age at T2DM</td>
<td></td>
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<tr>
<td></td>
<td>combinatio</td>
<td></td>
<td>with type 2 diabetes consecutively</td>
<td></td>
<td>diagnosis, male sex,</td>
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<td>n</td>
<td></td>
<td>attending hospital based diabetes</td>
<td></td>
<td>smoking, T2DM duration,</td>
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<td></td>
<td>clinics</td>
<td></td>
<td>HbA1c, antihyperglycemic</td>
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<td>treatment, triglycerides,</td>
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<td>HDL-C, BMI, eGFR, albuminuria,</td>
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<td></td>
<td></td>
<td>and retinopathy.</td>
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</tbody>
</table>
### Supp table 3. Characteristics of studies investigating the effect of metformin on other diseases of ageing

<table>
<thead>
<tr>
<th>Study</th>
<th>Exposure</th>
<th>Comparator</th>
<th>Population</th>
<th>Diabetes control adjustment</th>
<th>N/events and Follow up</th>
<th>Covariates adjusted for in multivariable analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Metformin compared to Non-Diabetic or General Population</strong></td>
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<tr>
<td>Geoghegan 2004</td>
<td>Use of metformin</td>
<td>Non use of metformin</td>
<td>United Kingdom. Case-control. All patients</td>
<td>N/a</td>
<td>NR</td>
<td>Matched by age, sex, general practice and duration of available data. Adjusted for mean annual consulting rates.</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>N/a</td>
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<tr>
<td>Vestergaard 2005</td>
<td>Use of metformin &lt;150 DDD (defined daily dose) Or Use of metformin 150-499 DDD Or Use of metformin ≥ 1,300 DDD</td>
<td>Never use of metformin</td>
<td>Denmark. Case-control. Any subjects.</td>
<td>N/a</td>
<td>NR</td>
<td>Matched for age and sex. Adjusted for type 1 diabetes, type 2 diabetes, insulin use, sulphonylurea use, other OAD drugs, prior fracture, anti-epileptic drugs, use of diuretics, use of anxiolytics and sedatives, neuroleptics, antidepressants, alcoholism, statins, non statin cholesterol lowering drugs, antihypertensives, myocardial infarction, stroke, days in bed in 1999, working or not, living with another person vs alone.</td>
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<tr>
<td><strong>Metformin compared to other diabetes therapies, controlling for diabetes management</strong></td>
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<tr>
<td>Hung 2013</td>
<td>Metformin use Or Metformin + Sulphonylurea use</td>
<td>Sulphonylurea use</td>
<td>USA. Cohort. Aged ≥ 18 years who filled an incident oral hypoglycemic drug prescription during the study period, excluding those with severe medical conditions, poor kidney function or heart failure</td>
<td>HbA1c over time</td>
<td>Glomerular filtration rate or end stage renal disease events</td>
<td>Age, sex, baseline creatinine, baseline blood pressure, history of hypertension, history of cardiovascular disease, baseline HbA1c, baseline BMI, the use of ACEI or ARBs, diuretics, baseline number of medications, year of cohort entry, number of outpatient visits, history of hospitalisation at baseline, marital status.</td>
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<tr>
<td>Lin 2015</td>
<td>Metformin use (more than 110g)</td>
<td>Non-use of metformin</td>
<td>USA. Cohort. Aged 40 years or older enrolled for more</td>
<td>HbA1c over time</td>
<td>Open angle glaucoma</td>
<td>Age, sex, race, socioeconomic factors, geographic region,</td>
</tr>
</tbody>
</table>
within a 2 year window) than 2 consecutive years, diagnosed with diabetes within 2 years of being enrolled. Overall= 150,016/ 5,893 Mean 52.8-63.3 months cormorbid ocular disease, comorbid medical conditions, type of diabetes, Charlson comorbidity index, cataract surgery, retina surgery, diabetic control measured by HbA1c over time.

<table>
<thead>
<tr>
<th>Masica 2013</th>
<th>Metformin use (ongoing prescription s)</th>
<th>Sulphonylurea use (ongoing prescriptions) Or THZ use</th>
<th>USA. Cohort. New users of oral anti diabetic medications.</th>
<th>HbA1c over time (quantified using AUC)</th>
<th>Glomerular filtration reduction or Proteinurea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metformin= 1,314/ NR Sulphonylurea= 209/NR TZDs= 103.</td>
<td>Overall 133 glomerular filtration events, 72 incidences of proteinurea</td>
<td>Mean 1.5-2.0 years Age, sex, race, HbA1c, renal function, congestive heart failure, coronary heart disease, hypertension, BMI, diabetes diagnosis year</td>
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</table>

<table>
<thead>
<tr>
<th>Ng 2014</th>
<th>Metformin taken in the year before baseline Or Metformin taken for 6 years or less before baseline Or Metformin taken for more than 6 years before baseline</th>
<th>No metformin use in the year before baseline</th>
<th>Singapore. Cohort. People with a diagnosis of diabetes using anti-diabetic medications.</th>
<th>Fasting blood glucose at baseline</th>
<th>Cognitive impairment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metformin= 204/ NR Non-users= 161/NR</td>
<td>Mean 3.9 years</td>
<td>Age, gender, education, other anti diabetic medication use, fasting blood glucose, duration of diabetes, BMI, hypertension, cardiovascular illness or stroke, other medical comorbidities, eGFR, GDS score, APOE-ε4 allele, and duration of follow up.</td>
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</table>

| Kahn 2008 | Initially 500 mg metformin titrated to the maximum effective daily dose (1g twice daily) | Initially 4 mg rosiglitazone titrated to the maximum effective daily dose (4mg twice daily) | USA. Randomised controlled trial. People with type 2 diabetes (diagnosed within 3 years) naive to oral hypoglycemic drugs, aged 30-75, fasting plasma glucose 126 to 180mg/dl. Exclusion criteria were clinically significant liver disease, renal impairment, a history of lactic acidosis, unstable or severe angina, | Current A1C | Fracture Metformin= 1454/ 59, Rosiglitazone = 1456/ 92 Mean 3.3-4.0 years | Current values of weight, serum creatinine, hemotocrit, calcium, A1C, and waist circumference. |

Fracture Metformin= 1454/ 59, Rosiglitazone = 1456/ 92 Mean 3.3-4.0 years | Current values of weight, serum creatinine, hemotocrit, calcium, A1C, and waist circumference. |
known congestive heart failure, uncontrolled hypertension, or chronic disease requiring periodic or intermittent treatment with oral or intravenous corticosteroids or continuous use of inhaled corticosteroids.