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Meta-analysis: Efficacy of Pioglitazone and Metformin in the Treatment of Nonalcoholic Steatohepatitis

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An abstract of
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Abstract

Meta-analysis: Efficacy of Pioglitazone and Metformin in the Treatment of Nonalcoholic Steatohepatitis

By Henry Olejeme

Background: This meta-analysis investigates the efficacy of pioglitazone and metformin in the treatment of nonalcoholic steatohepatitis (NASH). A recent meta-analysis demonstrated efficacy of pioglitazone, but found no benefit with metformin in the treatment of NASH. The outcome measures in that study were expressed as any improvement (yes versus no) regardless of magnitude. Using a different methodology that considers continuous outcome measures we set out to determine if the results of our meta-analysis and the one published previously arrive at similar or different conclusions.

Methods: Multiple online databases were searched and reports of randomized controlled trials reviewed. The summary results were expressed as meta-differences for continuous endpoints of histology and liver function tests. All summary measures of effect were calculated using fixed and random effects models accompanied by a corresponding 95% confidence interval (CI) and a test for heterogeneity.

Results: None of the random effects models demonstrated a statistically significant departure from the null for pioglitazone, ALT (meta-DD, 3.17; 95% CI: -17.46, 23.81), AST (meta-DD, -7.03; 95% CI: -28.71, 14.65) or metformin, ALT (meta-DD, 6.02; 95% CI: -8.84, 20.87), AST (meta-DD, 5.20; 95% CI: -5.34, 15.73). Similarly, pioglitazone use was not associated with improvement in any of the histological parameters: steatosis (meta-DD, -0.03; 95% CI: -0.62, 0.55), inflammation (meta-DD, -0.16; 95% CI: -0.62, 0.30), ballooning (meta-DD, 0.04; 95% CI: -0.33, 0.41), fibrosis (meta-DD, 0.17; 95% CI: -0.15, 0.49).

Conclusions: Unlike the previously published meta-analysis, our results do not favor the use of pioglitazone or metformin for the treatment of NASH. The different conclusions drawn from the two meta-analyses are likely attributable to variations in analytic techniques.

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Introduction

Nonalcoholic fatty liver disease (NAFLD) encompasses a spectrum of conditions.[1] Whereas the majority of patients with NAFLD have simple steatosis that has a benign course, about 20% of patients will have a more severe form known as non-alcoholic steatohepatitis (NASH), a histological diagnosis that consists of steatosis, hepatocyte ballooning and lobular inflammation with or without fibrosis. Compared with patients with simple steatosis, those with NASH are more likely to progress to cirrhosis, liver failure, or hepatocellular carcinoma.[2]

NASH is becoming the leading cause of chronic liver disease and a major health issue owing to its close association with the worldwide epidemics of obesity and diabetes.[3] It has become the most common cause of abnormal liver tests in the United States and an important cause of liver cirrhosis.[4] This condition is projected to be the leading cause of liver transplantation by 2020.[5] NASH is also associated with an increased risk of cardiovascular mortality and type 2 diabetes mellitus.[6] Accordingly, the treatment of NASH is now considered a relevant issue in clinical hepatology and several therapeutic approached have been tested in pilot, controlled and uncontrolled studies.[7]

Insulin resistance is an almost universal finding in primary NASH. It is the main driving force behind excessive fat accumulation in the liver but may also play a role in the initiation and perpetuation of steatohepatitis and fibrosis progression.[8-10] Although no pharmacologic therapy has been conclusively proven to be effective for the treatment of NASH,[11] the importance of insulin resistance in the pathophysiology of this condition may warrant the use of oral hypoglycemic agents. Thiazolidinediones (especially pioglitazone) are the most well-studied compounds to date for the treatment of NASH.[4] The insulin sensitizing, anti-inflammatory, and antifibrotic properties of thiazolidinediones support their use.[12] Studies involving pioglitazone

have shown the most promise. A meta-analysis of randomized controlled trials (RCT) of thiazolidinediones (TZD) for the treatment of NASH showed that TZDs improve steatosis and inflammation.[5] The studies conducted to date, however, were limited by size and duration.[5]

Other insulin sensitizers such as metformin have also been evaluated in RCTs for the treatment of NASH. A recent meta-analysis concluded that 6-12 months of metformin plus lifestyle intervention did not improve liver histology or aminotransferases compared with lifestyle intervention alone.[5]

Meta-analyses, a subset of systematic reviews, employ formal statistical methodology to combine the results of several studies in an effort to more precisely quantify the benefits and risk of medical interventions. Evidence based medical practice has relied on meta-analyses to overcome the limitations of individual small trials that may not be powered enough to provide robust conclusions about relatively small effect sizes.[13] Systemic reviews or meta-analysis are supposed to be a reflection of the state of science that allows physicians to draw conclusions. Meta-analyses however, are subject to uncertainty and disagreements, which can be influenced by inclusions and exclusions, opinions, and quality of paper analytic techniques.[14]

The purpose of this paper is two-fold. First we aim to revisit the literature by performing a meta-analysis of the RCTs evaluating efficacy of pioglitazone and metformin in treatment of NASH. In addition our goal is to determine if the two meta-analyses – the current and the one published previously [5]– arrive at similar or different conclusions. The latter goal will allow us to estimate to what extent the results may be affected by inclusion/exclusion, criteria, judgment about individual study quality and choice of analytic methods.

Methods

Literature search and selection criteria

A computer-assisted search was conducted to identify potentially relevant publications in PubMed and the Cochrane Central Register of Controlled Trials. The databases were searched from their inception until 22 February 2012. Indexing terms included non-alcoholic steatohepatitis or non-alcoholic steatohepatitis or NASH in combination with Pioglitazone, non-alcoholic steatohepatitis or non-alcoholic steatohepatitis or NASH in combination with Metformin or non-alcoholic steatohepatitis or non-alcoholic steatohepatitis or NASH in combination with insulin sensitizers. A manual review of the bibliographies of seminal primary and review articles was also performed to identify additional studies.

Potentially relevant studies were selected based on the following inclusion criteria: (i) randomized controlled trials in patients with NASH using metformin or pioglitazone); (ii) diagnosis of NASH determined by histology; (iii) well-defined treatment outcomes, that include one or more of the following: changes in serum ALT, AST, bilirubin or liver histology parameters related to NASH. (iv) adult patients. Studies in which NASH was diagnosed based on elevated transaminases or abnormal imaging studies (ultrasound, CT scan, magnetic resonance imaging) without histological confirmation were excluded. Case reports or series were excluded as were review articles. A map of the literature search and selection process is detailed in **Figure 1.**

Data extraction

Eligible articles were reviewed by the author. For each study, the following data were obtained: (i) Study: year, design, size; (ii) Treatment: dose, frequency, duration; (iii) Patients: number,

mean age, gender; (iv) Laboratory tests (baseline and end of study): AST, ALT, bilirubin, GGT, alkaline phosphatase; and (v) Histology (baseline and end of study): steatosis grade, hepatocyte ballooning score, lobular inflammation score and fibrosis stage.

Primary and secondary outcomes

The primary outcomes for this meta-analysis were histological responses to treatment, specifically changes in: (i) steatosis grade (0–3), (ii) hepatocyte ballooning score (0– 2), (iii) lobular inflammation score (0–3), or (iv) fibrosis stage (0–4). The secondary outcomes included (i) biochemical response, measured as change in ALT, AST, and Bilirubin.

Quality assessment

The studies included in the meta-analysis were assessed for quality based on the Jadad 3-point scale [Was the study described as randomized (this includes words, such as randomly, random and randomization)? Yes = 1, No = 0. Was the method used to generate the sequence of randomization described and appropriate (table of random numbers, computer-generated, etc.)? Yes = 1, No = 0. Was the study described as double-blind? Yes = 1, No = 0. Was the method of blinding appropriate (complete versus incomplete blinding or possibility that blinding could have been broken)? Yes=1, No=0. Was there an account of all patients in the trial (including withdrawals from study or death)? Yes=1, No=0].[15] The quality assessment scheme for this study is shown in **Table 1**.

Study quality was further assessed using the Cochrane Collaboration's tool for assessing risk of bias.[14] As opposed to the Jadad scale that utilize scales or check lists, this tool utilizes domain-based evaluation, which focuses on criteria that are more highly associated with internal validity in RCTs.[14] Quality assessment was based on the following domains: (i) randomization, (ii) allocation concealment, (iii) blinding of participants, personnel, and outcome assessors, (iv)

completeness of outcome data, (v) unbiased outcome reporting and (vi) lack of other sources of bias. The quality assessment for this study using this tool is described under the comment section of **Table 2.**

Statistical Analysis

The before-and-after differences in outcome measures were calculated for the intervention and the comparison groups separately and the difference of differences was then estimated using the equation: (treatment mean at baseline – treatment mean at the end of the study) – (control mean at baseline – control mean at the end of the study). Using this approach if the biochemical or histological response to treatment improved to a greater extent in the intervention compared with the control group, the difference would assume a positive value. Difference in means for each outcome for intervention and control group was either directly obtained from the study results or calculated by using the data provided in the article.

All measures of effect (difference of differences, henceforth denoted as DD) were accompanied by the corresponding measure of variance. Those were usually calculated based on the p-values, which were used to obtain Z scores, then back calculating the standard deviations (SDs) and the corresponding estimates of variance. The p-value for the change from baseline between the intervention and control group was reported by most of the studies.[11, 12, 16, 17 18] In two of the studies this was not reported[19, 20] and the p-value was derived using the following approach. First, SD for the before-and-after difference in each group was calculated as square root of [(SD pre-intervention mean)^2 + (SD post-intervention mean)^2]; and then the DD was calculated and the corresponding p-value was obtained using the two sample independent t-test in the OpenEpi software. Thus, prior to inclusion in the meta-analysis each study provided a measure of effect (DD) and the corresponding variance.

All meta-analyses were performed using EpiSheet, a spreadsheet-based analytical package.[21] The summary results (meta-DD) were calculated using both random and fixed effects models accompanied by a corresponding 95% confidence interval (CI), and a test for heterogeneity.[22]

Results

Study description

Seven trials were included in our meta-analysis; four of those investigated the use of pioglitazone and the remaining three assessed the efficacy of metformin. The four pioglitazone studies had treatment durations of 6 months,[11, 20] 12 months,[12] and 24 months.[16] The metformin trials used dosages ranging from 500 mg to 1700 mg daily. Duration of treatment was 6 months in one study and 12 months in the other two.[17-19] All trials excluded patients with benign steatosis and enrolled only patients with NASH. Characteristics of the seven studies included in the meta-analysis are presented in **Table 2.**

Meta-analysis: changes in liver function tests.

The results of the meta-analysis of studies evaluating the effect of oral hypoglycemic agents on ALT and AST are summarized in **Tables 3 and 4** respectively. Pooled results of studies of metformin[17-19] but not pioglitazone[11, 12, 16, 20] use demonstrated an improvement in ALT (meta-DD, 4.74; 95% CI: 1.14, 8.33) and AST (meta-DD, 5.23; 95% CI: 2.77, 7.69), but only in the fixed effects models. By contrast none of the random effects models demonstrated a statistically significant departure from the null. Sub-analyses limited to studies of the highest quality,[11, 16, 18] each with a Jadad score of 4 out of 5, did not provide any evidence to support improvement in ALT or AST following administration of glycemia lowering drugs. For the meta-analyses and sub-analyses, all p-values for homogeneity were <0.005 suggesting that the studies were quite heterogeneous, and supporting the use of random effects models.

As shown in **Table 5**, pooled results of the three studies evaluating the effect of pioglitazone on bilirubin levels,[12, 16, 20] demonstrated that pioglitazone use was associated with a small but statistically significant increase in bilirubin level in the fixed model (meta-DD, -0.13; 95% CI: -0.24, -0.02). By contrast the corresponding result for the random effects model was not

statistically significant (meta-DD, -0.13; 95% CI: -0.35, 0.09). The p for homogeneity in this analysis was 0.109.

Meta-analysis: changes in liver histology.

Histological outcomes were not analyzed for the metformin trials because one of those studies did not report results of histological outcomes other than fibrosis,[17] one reported median rather than mean scores,[19] and the third used a different scoring scale.[18] For this reason, **Table 6** summarizes the pooled treatment effect of only pioglitazone. Three of the four pioglitazone studies had overall quality scores of 4,[11, 16, 20] and one had a score of 3.[12] Pooled results from these four trials using both fixed and random effects demonstrate that pioglitazone use was not associated with improvement in any of the histological parameters including steatosis, lobular inflammation, ballooning degeneration or fibrosis. Across all parameters, p for homogeneity was <0.005 suggesting that the studies were quite heterogeneous.

Discussion

Our analysis highlights the variations in meta-analysis results that can occur due to differences in methods. We found no significant effect of hypoglycemic drugs on liver function test result and no benefit of pioglitazone in improving the histological parameters of NASH. In contrast, a previous meta-analysis by Musso et al that addressed the same research question and included the same four studies[11, 12, 16, 20] of pioglitazone and one RCT of another TZD, rosiglitazone[23], arrived at a different conclusion.[5] In their meta-analysis, pooled results of RCTs showed that TZD's improved histological steatosis and inflammation but not fibrosis.[5] In contrast to our meta-analysis, heterogeneity was low for all of their assessed outcomes, suggesting a consistent drug effect size across studies.[5] The variation seen in the results of these two meta-analysis is likely attributable to the differences in analytic techniques used. In our meta-analysis, the effect was expressed as difference of differences in means (DD) for each outcome for intervention and control groups, whereas Musso and colleagues used dichotomous variables for any improvement (Yes versus No) that did not take into consideration the magnitude of absolute change in the parameters of interest.

Similar to the findings, the meta-analysis by Musso et al[5] did not find any statistically significant improvement in ALT or AST for metformin. It is important to note, however, that they calculated pooled effects using only random effects models, whereas we used both the random and the fixed effects method. In our study the fixed effects model demonstrated a statically significant improvement in ALT and AST which was not seen when the random effects assumptions were used. However in the presence of substantial heterogeneity a random effects model is considered more appropriate.[24]

Musso et al[5] included an additional study of metformin in patients with NAFLD, purposefully excluded from our analysis because this study was not strictly limited to patients with NASH, our

primary focus. The analytic techniques used by Musso et al in this case, were similar to ours in the use of continuous variables and DD.

The approach to quality assessment in systematic reviews is inconsistent and often debated.[25] Whereas some authors accept and support the use of formal quality scores of individual studies, others consider them unscientific and do not recommend their use. [27, 28,29] The Jadad scale that we used in this study is not without its critics, who have charged that it is over-simplistic, placing too much emphasis on blinding,[26] and can show low consistency between different raters.[27] These criticisms notwithstanding, our sub-analysis limited to studies that received the highest quality score did not affect the results.

Cochrane Collaboration Risk of Bias Tool (CCRBT) developed in 2008 represents a shift away from the assessment of methodological quality of studies using scales, and towards an evaluation of the *risk of bias* in the results of each study through a domain-based evaluation.[28] This distinction between methodological quality and risk of bias, was made in recognition of the fact that a study may be performed to the highest quality standards, but still have an important risk of bias.[28] The CCRBT evaluates six different domains and provides guidelines to score each item.[28] The CCRBT was used to assess quality in the reference meta-analysis.[5] The studies included in our sub-analysis when compared to the reference, similarly received very high scores and were categorized as high quality (HQ).

The notable strengths of this meta-analysis, include the strict inclusion criteria (RCTs with a histological definition of NASH) and the assessment of both histological and biochemical outcomes. The treatment duration of the majority of the studies was at least 6 months or greater. Most studies were of high quality, there were few dropouts in the trials, and follow-up was adequate.

There were also several limitations. Notably, relatively few studies were available for inclusion in our meta-analysis. P-values were not always provided in some of the studies necessitating the use of assumptions to approximate p-values. The trials included in our review were heterogeneous not only with respect to their results, but also with respect to methods used. Some of the trials enforced strict diet and exercise regimens in addition to the treatment, while others did not incorporate any lifestyle modification. With the exception of ALT and AST, there was little consistency in reporting of liver function tests (LFTs), and for this reason our meta-analysis was limited to relatively few LFTs.

In the assessment of histological parameters, there were variations in score ranges and systems used for measuring steatosis, hepatocyte ballooning, lobular inflammation and fibrosis, making it difficult to compare the degree of changes in these parameters across all studies. In addition, the recorded results varied extensively, with some studies only reporting the percentage of patients with an improvement in score, while others reporting the actual change in score. Additional calculations had to be performed in order to approximate the scoring system and this could have influenced our results. As with all studies that utilize histological endpoints, our findings are limited by the inherent sampling variability with resultant inaccuracies sometimes associated with liver biopsies.[29]

In conclusion, the disagreement of our results with those of the previous meta-analysis by Musso et al illustrates how different review methodology may affect interpretation of evidence. Although our meta-analysis does not favor the use of glycemia lowering medications for the treatment of NASH, our results need to be interpreted with caution due to the limitations of our study and due to shortcomings of individual papers included in the review. Given the limited number of studies that met our inclusion criteria, additional studies with similar endpoints are

needed to adequately assess the effect of pioglitazone and metformin in NASH. Future trials of NASH would benefit from standardizing study design, treatment outcomes and histological scoring.

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Figure and Tables

Figure 1: Summary of the literature search

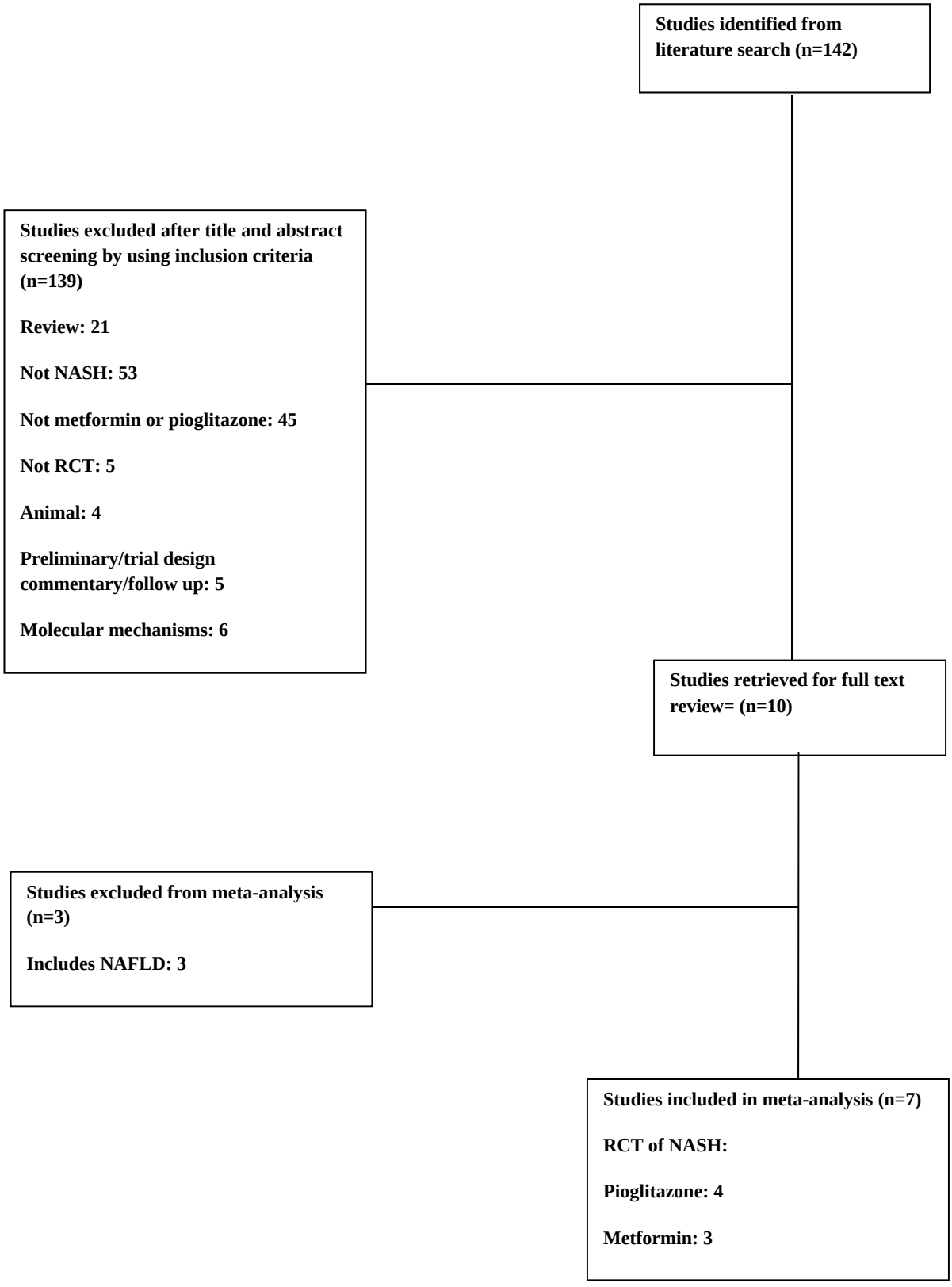

Table 1: Jadad Scale for evaluating individual study quality

Item	Maximum Points	Description	Sanyal 2010	Aithal 2008	Belfort 2006	Sanyal 2004	Shields 2009	Uygun 2004	Idilman 2008
Randomization	**2**	**1 point if randomization is mentioned**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
		1 additional point if the method of randomization is appropriate	**0**	**0**	**1**	**1**	**1**	**1**	**0**
		Deduct 1 point if the method of randomization is inappropriate (minimum 0)	**0**	**0**	**0**	**-1**	**0**	**0**	**0**
Blinding	**2**	**1 point if blinding is mentioned**	**1**	**1**	**1**	**1**	**1**	**0**	**0**
		1 additional point if the method of blinding is appropriate	**1**	**0**	**0**	**0**	**0**	**0**	**0**
		Deduct 1 point if the method of blinding is inappropriate (minimum 0)	**0**	**0**	**0**	**-1**	**0**	**0**	**0**
An account of all patients	**1**	**The fate of all patients in the trial is known. If there are no data the reason is stated.**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
TOTAL SCORE	**5**		**4**	**3**	**4**	**2**	**4**	**3**	**2**

Table 2: Characteristics of studies included in the meta-analysis

Author	Duration (months)	Age (yr)	M%	Dose	Study size		Design Jadad Score	Comments	Results LFT	Results Histology
					T	C				
Pioglitazone										
Sanyal 2010	24	47	41	30 mg	80	83	RCT/4	RSG: Yes AC: Yes Blinding of participants: Yes Blinding of personnel: No (local pathologist for eligibility), Yes (pathologists for final analysis) Blinding of OA: Yes Drop out T: 14 Drop out C: 12 Medication compliance: NR Missing biopsy T: 10 Missing biopsy C: 11 ***missing biopsy imputed as no improvement*** Strengths: RCT, larger sample size and duration relative to prior studies, multicenter Weaknesses: High inter-observer variability in histologic	Improved	Improved steatosis and inflammation (NS)

								assessment of pioglitazone group, unblinded local pathological review for eligibility. Not generalizable to diabetics and cirrhotics who were excluded from study.		
Aithal 2008	12	52	70	30 mg	37	37	RCT/3	RSG: Yes AC: Yes Blinding of participants: Yes Blinding of personnel: Yes Blinding of OA: Yes Drop out T: 6 Drop out C: 7 Medication compliance: 99% in all but 1 (97%) Missing biopsy T: 6 Missing biopsy C: 7 ***missing biopsy excluded in analysis*** Strengths: RCT with placebo arm, multicenter Weaknesses: small sample size and short study duration	Improved (NS)	Improved hepatocellular injury and fibrosis; steatosis and inflammation improved (NS)
Belfort 2006	6	51	54	45 mg	26	21	RCT/4	RSG: Yes AC: Yes Blinding of participants: Yes Blinding of personnel:	Improved	Improved steatosis and inflammation. Fibrosis improved (NS)

								Yes Blinding of OA: Yes Drop out T: 3 Drop out C: 4 Medication compliance: 95% in all but 1 (50%) Missing biopsy T: 3 Missing biopsy C: 4 ***missing biopsy excluded in analysis*** Strengths: RCT with placebo arm, multicenter, good intra and inter-observer histologic reading agreement Weaknesses: not generalizable to non diabetics since study consisted primarily of diabetics or impaired GTT, small sample size and short study duration		
Sanyal 2004	6	47	60	30 mg	10	10	RCT/2	RSG: Yes AC: No Blinding of participants: No Blinding of personnel: No Blinding of OA: Yes Drop out T: 2 Drop out C: 0 Medication compliance: NR	Improved	Improved steatosis and inflammation. Fibrosis improved (NS)

								Missing biopsy T: 2 Missing biopsy C: 0 ***missing biopsy excluded in analysis*** Strengths: RCT Weaknesses: small sample size and short study duration, poorly blinded		
Metformin										
Shields 2009	12	50	89	500 - 1000 mg	9	10	RCT/4	RSG: Yes AC: Yes Blinding of participants: Yes Blinding of personnel: Yes Blinding of OA: Yes Drop out T: 0 Drop out C: 3 Medication compliance: NR Missing biopsy T: 0 Missing biopsy C: 3 ***data in dropout included in final intention-to-treat analysis*** Strengths: RCT with placebo arm, Weaknesses: small sample size and short study duration, M:F ratio of 8:1 in treatment group vs equal ratio in placebo, low dose of metformin	Improved (NS)	Improved (NS)

Uygun 2004	6	40	65	1.7 mg	17	17	RCT/3	RSG: Yes AC: No Blinding of participants: No Blinding of personnel: No Blinding of OA: No Drop out T: 1 Drop out C: 1 Medication compliance: NR Missing biopsy T: 4 Missing biopsy C: 7 ***missing biopsy excluded in analysis*** Strengths: RCT Weaknesses: unblinded, no placebo group, small sample size and short study duration, no post treatment biopsy in some	Improved (NS)	Improved necroinflammation (NS)
Idilman 2008	12	48	44 ##	1.7 mg	24	25	RCT/2	RSG: Yes AC: No Blinding of participants: No Blinding of personnel: No Blinding of OA: Yes Drop out T: 0 Drop out C: 0 ***second treatment arm with rosiglitazone had 1 dropout****	Improved (NS)	Improved steatosis

								Medication compliance: NR Missing biopsy T: 14 Missing biopsy C: 17 ***missing biopsy excluded in analysis*** Strengths: RCT Weaknesses: poorly blinded, no placebo group, small sample size and short study duration, large numbers of missing biopsies (post treatment)		

KEY:

RSG: Random Sequence Generation
AC: Allocation Concealment
OA: Outcome Assessment
T: Treatment group
C: Control group
RCT: Randomized Controlled Trial
GTT: Glucose Tolerance Test
NS: Nonsignificant
##: Male percentage for metformin and rosiglitazone group
NR: Not Reported

Table 3: Meta-analysis of studies evaluating effect of hypoglycemic agents on ALT results

Studies evaluating effect of hypoglycemic drugs on ALT levels			
Author	DD	Variance	p-value
Sanyal 2010	-20.7	39.59	0.001
Aithal 2008	30.8	139.26	0.009
Belfort 2006	18.0	29.93	0.001
Sanyal 2004	-10.0	22.25	0.034
Idilman, 2008	8.00	6.41	<0.001
Uygun 2004	19.70	44.00	<0.001
Shields, 2009	-19.20	137.06	0.100
Meta-analysis results: all studies (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	3.60	-8.35	15.55
Fixed Effects	4.74	1.14	8.33
Meta-analysis results: pioglitazone only (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	3.17	-17.46	23.81
Fixed Effects	-1.35	-7.24	4.54
Meta-analysis results: metformin only (p for homogeneity 0.014)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	6.02	-8.84	20.87
Fixed Effects	8.36	3.82	12.91
Meta-analysis results: studies of highest quality (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-6.60	-35.29	22.10
Fixed Effects	-0.93	-8.57	6.70

Table 4: Meta-analysis of studies evaluating effect of hypoglycemic agents on AST results

Studies evaluating effect of hypoglycemic drugs on AST levels			
Author	DD	Variance	p-value
Sanyal 2010	-16.6	25.46	0.001
Belfort 2006	10.0	23.80	0.040
Sanyal 2004	-20.0	251.95	0.224
Idilman 2008	6.00	2.12	<0.001
Uygun 2004	15.30	15.47	<0.001
Shields 2009	-14.40	77.10	0.100
Meta-analysis results: all studies (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-0.07	-9.77	9.64
Fixed Effects	5.23	2.77	7.69
Meta-analysis results: pioglitazone only (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-7.03	-28.71	14.65
Fixed Effects	-3.65	-10.36	3.06
Meta-analysis results: metformin only (p for homogeneity 0.005)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	5.20	-5.34	15.73
Fixed Effects	6.61	3.97	9.26
Meta-analysis results: studies of highest quality p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-6.54	-25.53	12.45
Fixed Effects	-4.44	-10.82	1.94

Table 5: Meta-analysis of studies evaluating effect of pioglitazone on bilirubin results

Studies evaluating effect of pioglitazone on bilirubin levels			
Author	DD	Variance	p-value
Sanyal 2010	-0.10	0.00	0.070
Aithal 2008	4.00	4.53	0.06
Sanyal 2004	-0.20	0.01	0.05
Meta-analysis results: all studies (p for homogeneity 0.109)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-0.13	-0.35	0.09
Fixed Effects	-0.13	-0.24	-0.02

Table 6: Meta-analysis of studies evaluating effect of pioglitazone on liver histology parameters

Studies evaluating effect of pioglitazone on steatosis

Author	DD	Variance	p-value
Sanyal 2010	-0.70	0.05	<0.001
Aithal 2008	0.19	0.02	0.190
Belfort 2006	0.80	0.07	<0.001
Sanyal 2004	-0.40	0.06	0.100
Meta-analysis results (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-0.03	-0.62	0.55
Fixed Effects	-0.02	-0.22	0.17

Studies evaluating effect of pioglitazone on inflammation

Author	DD	Variance	p-value
Sanyal 2010	-0.50	0.02	<0.001
Aithal 2008	0.07	0.00	0.250
Belfort 2006	0.53	0.04	0.010
Sanyal 2004	-0.80	0.06	<0.001
Meta-analysis results (p for homogeneity <0.001)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	-0.16	-0.62	0.30
Fixed Effects	-0.01	-0.11	0.10

Studies evaluating effect of pioglitazone on ballooning

Author	DD	Variance	p-value
Sanyal 2010	-0.20	0.01	0.010
Aithal 2008	0.33	0.01	0.010
Belfort 2006	0.54	0.05	0.020
Sanyal 2004	-0.40	0.02	<0.001
Meta-analysis results (p for homogeneity <0.001)			
Model	meta-DD	95% CI lower	95% CI upper
Random Effects	0.04	-0.33	0.41
Fixed Effects	-0.07	-0.18	0.04

Table 6 (continued): Meta-analysis of studies evaluating effect of pioglitazone on liver histology parameters
Studies evaluating effect of pioglitazone on fibrosis

Author	DD	Variance	p-value
Sanyal 2010	-0.30	0.03	0.100
Aithal 2008	0.42	0.05	0.050
Belfort 2006	0.46	0.07	0.080
Sanyal 2004	0.20	0.01	0.100
Meta-analysis results (p for homogeneity 0.027)			
Model	Meta-DD	95% CI lower	95% CI upper
Random Effects	0.17	-0.15	0.49
Fixed Effects	0.15	-0.02	0.32