# Effect of fetal bovine serum concentration towards vero cells growth on culture in DMEM medium

*by* .Abdullah _TURNITIN

**Submission date:** 05-Jun-2024 02:28PM (UTC+1000)
**Submission ID:** 2390776962
**File name:** ween-Type-2-Diabetes-Mellitus-with-and-without-Complications.pdf (364.97K)
**Word count:** 5346
**Character count:** 28419

ORIGINAL PAPERS

# Differences in AGEs and hs-CRP between Type 2 Diabetes Mellitus with and without Complications

Alfino Validita SIDIQ[1], Yetti HERNANINGSIH[2,4], Puspa WARDHANI[2,4], Soebagijo Adi SOELISTIJO[3,4]

## Abstract

**Background:** Diabetes mellitus is a metabolic disease that can lead to macrovascular and microvascular complications that can cause death and reduce the quality of life. The level of serum AGEs increases with diabetes or a state of hyperglycemia. Increased AGEs in patients with diabetes mellitus can increase the risk of macrovascular and microvascular complications and increase the formation of hs-CRP. This study aims to assess the differences in AGEs and hs-CRP between type 2 diabetes mellitus patients with and without complications and the relationship between AGEs and hs-CRP.

**Methods:** A cross-sectional study was conducted with 30 subjects of type 2 diabetes mellitus with complications and 30 subjects of diabetes mellitus without complications. Data were collected from September to December 2022. AGEs were measured using ELISA and hs-CRP using the turbidimetric method. Data were statistically processed to determine the difference and relationship between AGEs and hs-CRP.

**Results:** AGEs and hs-CRP values in patients with type 2 diabetes mellitus with complications were 0.671±0.194 ng/mL and 2.08(0.98-15.3) mg/L. In patients with uncomplicated type 2 diabetes 0.561±0.127 ng/mL and 1.69(0.22-9.25) mg/L. Differences in AGEs and hs-CRP between the two subject groups had significant results (AGEs p=0.012, hs-CRP p=0.038). The relationship between AGEs and hs-CRP in each subject group showed no significant results (With complications r=0.659 p=0.659, Without complications r=0.253 p=0.253).

**Conclusion:** There are differences in AGEs and hs-CRP values between patients with type 2 diabetes mellitus with and without complications and there is no relationship between AGEs and hs-CRP values in each subject

**Keywords:** Diabetes mellitus type 2, AGEs, hs-CRP, Complication.

## Rezumat

**Context:** Diabetul zaharat este o boală metabolică ce poate duce la complicații macro și microvasculare care pot cauza decesul și reduce calitatea vieții. Nivelul de AGE serice crește odată cu diabetul sau cu o stare de hiperglicemie. Creșterea AGEs la pacienții cu diabet zaharat poate crește riscul de complicații macrovasculare și microvasculare și poate crește formarea de hs-CRP. Acest studiu își propune să evalueze diferențele de AGE și hs-CRP între pacienții cu diabet zaharat de tip 2 cu și fără complicații și relația dintre AGE și hs-CRP.

**Metode:** A fost efectuat un studiu transversal cu 30 de subiecți de diabet zaharat de tip 2 cu complicații și 30 de subiecți de diabet zaharat fără complicații. Datele au fost colectate din septembrie până în decembrie 2022. AGEs au fost măsurate prin ELISA și hs-CRP prin metoda turbidimetrică. Datele au fost prelucrate statistic pentru a determina diferența și relația dintre AGEs și hs-CRP.

[1] Postgraduate Student, Basic Medical Science Master Program, Faculty of Medicine Universitas Airlangga, Surabaya, Indonesia
[2] Department of Clinical Pathology, Faculty of Medicine Airlangga University, Surabaya, Indonesia
[3] Department of Internal Medicine, Faculty of Medicine, Airlangga University, Surabaya, Indonesia.
[4] Dr Soetomo General Academic Hospital, Surabaya, Indonesia

**Corresponding author:**
**Yetti HERNANINGSIH,** Department of Clinical Pathology, Faculty of Medicine Airlangga University, Surabaya, Indonesia
**E-mail:** yetti-h@fk.unair.ac.id

**Rezultate:** Valorile AGEs și hs-CRP la pacienții cu diabet zaharat de tip 2 cu complicații au fost de 0,671±0,194 ng/mL și 2,08(0,98-15,3) mg/L. La pacienții cu diabet de tip 2 fără complicații 0,561±0,127 ng/mL și 1,69(0,22-9,25) mg/L. Diferențele de AGE și hs-CRP între cele două grupuri de subiecți au avut rezultate semnificative (AGE p=0,012, hs-CRP p=0,038). Relația dintre AGE și hs-CRP în fiecare grup de subiecți nu a avut rezultate semnificative (Cu complicații r=0,659 p=0,659, Fără complicații r=0,253 p=0,253).
**Concluzii:** Există diferențe în valorile AGEs și hs-CRP între pacienții cu diabet zaharat de tip 2 cu și fără complicații și nu există nicio relație între valorile AGEs și hs-CRP în fiecare grup de subiecți.
**Cuvinte cheie:** AGES și AGE: diabet zaharat tip 2, AGEs, hs-CRP, complicații.

## INTRODUCTION

Diabetes mellitus is a disease caused by metabolic disorders, often characterized by increased blood glucose levels in the body due to insulin secretion abnormalities. It is the most significant cause of death worldwide[1]. Type 2 diabetes mellitus is a health problem with an increasing prevalence worldwide. Indonesia is included in the top ten countries with the most estimated diabetes cases, collecting around 10.7 in 2019 and will jump to 13.7 million in 2030, then 16.6 million in 2045. The prevalence of diabetic complications, as demonstrated by multinational studies, recorded 27.2% of macrovascular complications and 53.5% of microvascular complications. In comparison, kidney complications accounted for 27.9%, eye disease 26.3%, diabetic foot 5.4%, and neuropathy 38.4%, significant causes of death in diabetic patients and reduced quality of life[2]. In 2012 diabetes caused 1.5 million deaths and 2.2 million deaths in patients with high glucose 43% of these 3.7 million deaths occurred before the age of 70 years, most of which were because of cardiovascular disease[3].

Advanced glycation end products (AGEs) belong to a heterogeneous group of compounds in general. AGEs are made by non-enzymatic condensation between the carbonyl group of reducing sugar and the free amine group of nucleic acid, protein, or lipid, resulting in a stable final product and irreversible[4]. AGEs can produce reactive oxygen species (ROS) as a source of inflammation. Serum levels of AGEs increase with age with diabetes or hyperglycemic states[5]. Patients with type 2 diabetes have hyperglycemia and low-grade inflammation, which may correlate with macrovascular diseases such as cardiovascular [6]. Receptors for advanced glycation end products (RAGE) are receptors for AGEs[5]. The presence of AGEs in the circulation that interacts with RAGE disrupts cellular properties, one of which is an increase in nuclear factor-kappa B (NF-kβ) transcription, which triggers an increase in interleukin-1 (IL-1), interleukin-6 (IL-6), tumor necrosis alpha (TNF-α) and causes ROS[7,8]. Increased inflammatory cytokines and abnormal glycated proteins can increase microvascular and macrovascular complications of diabetes[7].

C-Reactive protein (CRP) can also be called the acute phase produced by the liver. Increasing CRP levels in the body can be a marker of inflammation[9]. In diabetes, there is a mild increase in CRP levels. CRP examination can be taken by standard test and high-sensitivity CRP (hs-CRP). Measurements using hs-CRP can accurately detect low-grade inflammatory states[10]. Hyperglycemia can increase CRP production. Higher levels of hs-CRP can be found in diabetic patients and indicate that microvascular and macrovascular changes have occurred in these patients[11].

Increased AGEs in patients with diabetes mellitus can increase the risk of microvascular and macrovascular complications. Increased AGEs can also activate NF-kβ, which triggers an increase in IL-6 and TNF-α, a stimulator of hs-CRP[7,12,13]. Based on the explanation above, this study aimed to analyze the comparison of AGEs and hs-CRP in patients with type 2 diabetes mellitus with and without complications. This study aims to assess the differences in AGEs and hs-CRP between type 2 diabetes mellitus patients with and without complications and the relationship between AGEs and hs-CRP in each group.

## METHODS

### Study Design and Participant

This study was conducted in an analytical observational form with a cross-sectional by measuring serum AGEs and hs-CRP subject approach to determine whether there are differences between AGEs and hs-CRP in

subjects with type 2 diabetes mellitus with and without complications. Subjects were obtained based on patient medical record data from November - December 2022. The inclusion criteria of respondents were subjects with type 2 diabetes mellitus who were willing to be research subjects, over 18 years old, and had average body temperature. Exclusion criteria were not being an autoimmune disease, chronic infection, obesity, and taking inflammatory drugs and statins.

## ETHICS

The Ethical Committee of Rumah Sakit Universitas Airlangga Surabaya approved the study with the ethical permit number 096/KEP/2022. Informed written consent was obtained from all subjects participating in the study.

**Sample Size and Data Collection**

The total sample in this study was 60 subjects with type 2 diabetes mellitus, consisting of 30 subjects with type 2 diabetes mellitus with complications and 30 subjects with diabetes mellitus without complications. Interviewers collected respondent data and samples from research subjects.

**Assay AGEs Serum**

Venous blood samples from each subject were collected into plain tubes. The serum was separated by centrifugation at 3000-4000 rpm for 15 minutes. AGEs levels were determined at the Institute of Tropical Disease Airlangga University by ELISA *(Enzyme-linked immunosorbent assay)* Double antibody sandwich method using an ELISA Bio-Rad Laboratory with an ELISA kit Bioassay Technology Laboratory (Shanghai Korain Biotech Co., Ltd). The concentration of AGEs in ng/mL

**Assay hs-CRP**

Venous blood samples from each subject were collected into plain tubes. The serum was separated by centrifugation at 3000-4000 rpm for 15 minutes. Hs-CRP levels were determined at Airlangga University Hospital by the turbidimetric method using the TMS24 instrument (Tokyo Boeki Medisys) with Nanopia hs-CRP Reagent (Sekisui Medical Co., Ltd). This instrument automatically calculates the concentration of hs-CRP in mg/L.

**Statistical Analysis**

The data were processed using the statistical T-test for normally distributed data and Mann-Whitney for non-normally distributed to determine the difference in AGEs and hs-CRP values between the two subjects, and Spearman's correlation was used to determine the relationship between AGEs and hs-CRP because the data were not normally distributed with Jamovi 2.3.8.1 software assistance.

## RESULTS

### Characteristics of Research Subjects

A total of 60 subjects participated in this study, 30 with type 2 diabetes with complications and 30 with type 2 diabetes without complications.

**Table 1.** Demographics of the study population.

| Demographics of the study population. | T2DM with complication (n = 30) | T2DM without complication (n =30) | p-value |
|---|---|---|---|
| Age (Years) | 54,1 ± 7.47 | 55.3 ± 7.93 | 0,549[b] |
| Sex | | | 1,000[c] |
| Male | 16 (53,3%) | 16 (53,3%) | |
| Female | 14 (46,7 %) | 14 (46,7 %) | |
| Smoking | | | 0,405[c] |
| Yes | 8 (26,6%) | 11 (36,6%) | |
| No | 22 (73,4%) | 19 (63,4%) | |
| Duration of T2DM | | | 0,165[c] |
| < 5 years | 7 (23,3%) | 12 (40%) | |
| > 5 years | 23 (76,7%) | 18 (60%) | |
| BMI (kg/m$^2$) | | | 0,584[c] |
| Normal | 8 (26,6%) | 11 (36,6%) | |
| Overweight | 22 (73,4%) | 19 (63,4) % | |
| Sistole (mmHg) | 146 ± 22,2 | 139 ± 13,7 | 0,176[b] |
| Diastole (mmHg) | 83,5 ± 14,3 | 85,2 ± 9,24 | 0,586[b] |
| Fasting plasma glucose (mg/dl) | 131 (73 – 346) | 120 (72 - 329) | 0,544[a] |
| Postprandial plasma glucose (mg/dl) | 190 (125 – 426) | 180 (97 – 520) | 0,246[a] |

**[a] Mann Whitney, [b] Independent T test [c] Chi-Square, *significant p < 0,05**

**[d] Median (Min-Max), [e] Mean ± SD**

Table 1. illustrates that the research subjects have a mean age of 54.7 ± 7.67 years and the majority are male. The history of not smoking has a higher value than smoking in all study subjects. More research subjects have suffered from type 2 diabetes mellitus for more than five years from both categories. Subjects had more BMI values in the overweight category in diabetes with complications and without. The systolic value in type 2 diabetes research subjects with complications is higher. Diastolic values have almost the same mean values in both subject categories. The median value of fasting glucose was higher in patients with type 2 diabetes with complications compared to the median value of glucose in patients with type 2 diabetes mellitus without complications, as well as the median value of postprandial glucose.

### Analysis of differences in values of AGEs and hs-CRP

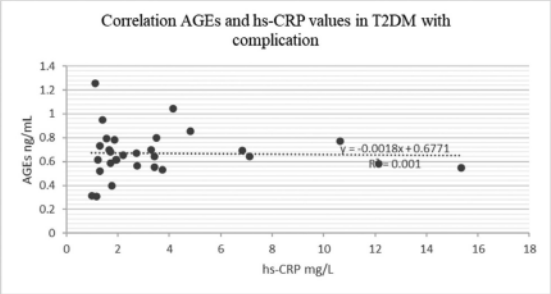
**Table.2** Differences in AGEs and hs-CRP values for T2DM with and without complications

| Examination | T2DM with complication (n=30) | T2DM without complication (n=30) | *p-Value* |
|---|---|---|---|
| AGEs (ng/mL) | 0,671 ± 0,194 | 0,561 ± 0,127 | 0,012[a] |
| hs-CRP (mg/L) | 2,08 (0,98 – 15,3) | 1,69 (0,22 – 9,25) | 0,038[b] |

**[*]a: Independent T-test, b: Mann-Whitney**

The difference between variables is said to be significant if p <0.05. Table 2 shows that the test results for both examinations obtained a value of <0.05, which means that there are differences in the two examination scores in the two categories of research subjects. AGEs and *hs*-CRP value examination results in type 2 diabetes mellitus subjects with complications have a higher value than those without complications.

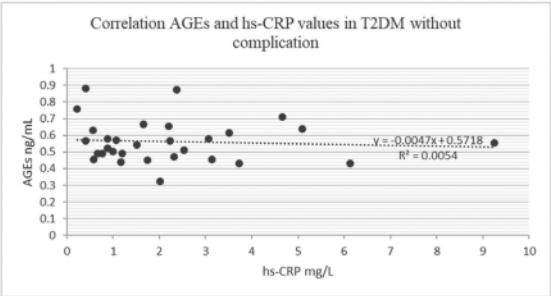
### Analysis of the correlation between values of AGEs and hs-CRP




*p-value = 0,659

**Figure 1.** Correlation between AGEs and hs-CRP T2DM with Complication.

Figure 1 shows no correlation between AGEs and hs-CRP values in patients with type 2 diabetes mellitus with complications *($r_s$ = 0.084)*.




*p-value = 0,253

**Figure 2.** Correlation between AGEs and hs-CRP T2DM without Complication.

Figure 2 shows no correlation between AGEs and hs-CRP values in subjects with type 2 diabetes mellitus without complications *($r_s$ = -0,255)*

## DISCUSSION

*Differences in AGEs values of the two categories of research subjects*

AGEs examination results with a higher average were found in type 2 diabetes mellitus subjects with complications compared to those without complications. This result is in line with the results obtained by Bansal et al., 2013 which stated that the AGEs value increased in the condition of type 2 diabetes subjects with complications compared to those without complications and have significant differences between the two categories. Poor glucose values can affect AGEs values and oxidative stress parameters, although some authors report that glycemic status in type 2 diabetes mellitus patients does not affect AGE levels.[14] in line with research conducted by Mostafa et al., 2007 which stated that increased AGEs were associated with the occurrence of kidney failure in diabetic patients and increased higher in patients with diabetes than without diabetes[15]. High AGEs values may be a predictor for diabetic nephropathy and correlate with the severity of nephropathy in diabetic patients. Higher levels of serum AGEs and inflammatory cytokines allow the development of microvascular and macrovascular complications in diabetes[14,16].

People with diabetes have increased formation of AGEs, AGE precursors, and oxidative stress due to increased circulating glucose[17] age and the irreversible nature of AGEs, the formation of AGEs occurs due to the glycation of proteins and lipids by glucose[18,19]. Advanced glycation Ends products which are non-enzymatic glycation products in lysine and arginine residues that accumulate in diabetic tissues, play an essential role in the pathogenesis of vascular complications in diabetics[14,18]. AGEs is an important molecule involved in various complications of diabetes. AGEs values increase blood circulation in patients with complicated diabetes mellitus[16].

The study's results stated that the presence of AGEs also plays a role in the emergence of type 2 diabetes, namely by making insulin resistance, β-cells become less functioning, and as a major player in diabetes complications.[19] AGEs in diabetic tissue correlated with diabetic complications[17]. AGEs can increase oxidative stress and increase the oxidation of low-density lipoprotein (LDL), which is a crucial player in the pathogenesis of heart disease; oxidised LDL acts as a ligand for RAGE, causing activation of several intracellular pathways that lead to hardening of the medial lining of

blood vessels which ultimately contribute to the pathophysiology of heart disease[20]. An increase in AGEs can be accompanied by an increase in inflammatory cytokines, which can cause various complications of conditions such as kidney, retina, nerves, and heart[21]. Arterial stiffness due to increased AGEs associated with micro-systemic inflammation can cause pressure fluctuations in the micro vessels of various organs, especially the kidneys. It can increase the risk of kidney failure.[20]

*Differences in hs-CRP values in the two categories of research subjects*

The results of the hs-CRP examination found statistically significant differences between the two categories, with higher average results found in type 2 diabetes mellitus subjects with complications compared to those without complications. This increase in CRP value is to research conducted by Aryan et al., 2018 which states that an increase in CRP value can increase the incidence of microvascular and macrovascular complications. Research by Babu & Joshi., 2017 states that there is an increase in hs-CRP accompanied by an increase in blood glucose in diabetic patients, along with Babu & Joshi., 2017 study by Tan et al., 2004. Increased CRP also occurs in people with diabetes compared to those without diabetes. High glucose levels can trigger microvascular changes and increased production of inflammatory factors, including CRP, IL-6, and TNF-α[22], although a minor increase occurs in diabetics[23].

The increase in hs-CRP in diabetes mellitus subjects with this complication could be due to an increase in proinflammatory cytokines such as TNF-α as a study conducted by Navarro et al. 2015 stated that there was an increase in TNF-α in diabetic neuropathy rat models. Proinflammatory cytokines are increased in diabetic patients indicating that inflammation may be related to diabetic complications[16,24]. CRP is also closely related to the risk of complications in people with diabetes mellitus (Gorska-Ciebiada et al., 2015; Volpe et al., 2018). Increased hs-CRP is closely related to the incidence of neuropathy, gangrene of the feet, diabetic retinopathy, and kidney failure (Aryan et al., 2018).

Previous studies have shown a close relationship between TNF-α and IL-6 in inflammation, which induces CRP transcription[27]. A meta-analysis study by Stanimirovic et al., 2022 showed that IL-6 mediates chronic inflammation in type 2 diabetes mellitus. High levels of IL-6 and TNF-α in diabetes mellitus have been demonstrated by several animal and human studies[27]. CRP formed in people with diabetes is partly due to the interaction between AGEs and RAGE modulating the formation of ROS through NADPH oxidation and mitochondria, which can increase pro-inflammatory cytokines and induce the formation of CRP.

The bond between AGE-RAGE activates several signals, including p21RAS, p44/p42 mitogen-activated protein kinase (MPAK), p113K-AKT, ERK1/2, JNK, p38, PKC, and NF-kβ activation. This signal becomes a regulator of many "response to injury" genes and ultimately results in the production of cytokines, chemokines, and other proinflammatory molecules that induce inflammation, apoptosis and proliferation, including molecules IL-6, TNF-α, IL-1β, and VCAM-1, which is regulated by AGEs[28].

*Correlation between AGEs and hs-CRP values in the two categories of research subjects*

The results of examining the relationship between AGEs and hs-CRP in the two categories of study subjects found that there was no statistically significant relationship, this is in accordance with the study of Pia de la et al 2012 which stated that AGEs values were not related to CRP in diabetic patients on anti-diabetic treatment orally or treated with insulin[29]. Research by Isa et al 2006 states that CRP values in people with uncontrolled diabetes also do not affect the decrease in AGEs values[30]. Factors affecting hs-CRP and AGEs results can be biased due to drugs that affect CRP and AGEs levels[31].

Research on the administration of metformin to patients with type 2 diabetes mellitus can significantly reduce the hs-CRP value compared to before receiving metformin. This decrease may be related to metformin therapy in patients with type 2 diabetes[32]. Reports obtained that metformin contains anti-inflammatory. Studies conducted in Canada show that metformin therapy can reduce mortality from heart disease by more than 45% in patients with type 2 diabetes mellitus.[33]

Metformin is an activator of adenosine monophosphate-activated protein kinase (AMPK), where the AMPK signal can suppress the activation of NF-kβ, thereby reducing the inflammatory response. Metformin can inhibit the inflammatory response generated by AGEs in macrophages through AMPK activation and suppression of RAGE/NF-kβ signals.[34] Metformin can also reduce hyperglycemia by increas-

ing peripheral insulin sensitivity. Only a tiny number of AGEs are formed in conditions with no hyperglycemia. Metformin can also increase glucose uptake in skeletal muscle and can inhibit glucose transport in the intestine so that it can reduce blood glucose levels when metformin is administered orally.[35]

Insulin can lower blood glucose and has an anti-inflammatory effect by suppressing NF-kβ activation; by decreasing the glucose value, the formation of AGEs is also decreasing. Insulin given to subjects without diabetes can suppress NF-kβ, a factor forming CRP. Injection of low doses of insulin to healthy patients can reduce ROS and CRP values in landmark studies[25,36] This is also consistent with a study conducted by Dwipayanan et al., 2017 that a decrease in CRP was found to be lower in the use of insulin compared to oral hypoglycemic drugs in patients with type 2 diabetes.

Smoking habits can increase CRP values, with minor increases[23] in research subjects, 31% of both categories with a smoking history. In the study, Gallus et al., 2018 stated that there was a significant relationship between hs-CRP and the duration and number of cigarettes consumed each day. This is due to the many free radicals entering the body. Advanced glycation Ends products can be sourced from within and outside, one of which is cigarettes which are also a source of AGEs from outside the body. Cigarettes are made by a process involving the formation of AGEs, and burning cigarettes can cause inhalation of AGE derivatives which can move into circulation[20,37]. Foods processed by frying and baking can form substantial AGEs, and food processing by enhancing taste can develop food-derived AGEs, also called glycotoxins. Advanced glycation Ends products with specific characteristics can be absorbed through food 10-30 per cent, although the metabolic process of absorption AGEs has not been thoroughly explained.[20]

### Study Limitations

The limitations of this study were using a small sample, not examining A1c to measure glycemic values in patients, not examining pro-inflammatory cytokines.

## CONCLUSION

This study found differences in AGEs and hs-CRP values between type 2 diabetes mellitus patients with and without complications. The values of AGEs and hs-CRP were higher in diabetic subjects with complications than those without complications. Furthermore, there is no correlation between AGEs and hs-CRP in patients with type 2 diabetes mellitus with complications and without complications.

### Acknowledgements

We thank the Faculty of Medicine Universitas Airlangga, Universitas Airlangga Hospital, and Institute of Tropical Diseases Universitas Airlangga. We also thank Mrs Atika, who helped analyze this study's statistics.

## References

1. Diabetes Care, "Standards of Medical Care in Diabetes," Diabetes Care, vol. 39, no. Supplement_1, pp. S4–S5, Jan. 2016, doi: 10.2337/dc16-s003.
2. D. Ardiany, A. Pranoto, S. A. Soelistijo, Libriansyah, and S. A. Widjaja, "Association between neutrophil–lymphocyte ratio on arterial stiffness in type-2 diabetes mellitus patients: a part of DiORS Study," Int J Diabetes Dev Ctries, vol. 42, no. 2, pp. 305–312, Apr. 2022, doi: 10.1007/s13410-021-00965-1.
3. R. J. Henning, "Type-2 diabetes mellitus and cardiovascular disease," Future Cardiology, vol. 14, no. 6. Future Medicine Ltd., pp. 491–509, Nov. 01, 2018. doi: 10.2217/fca-2018-0045.
4. A. Twarda-clapa, A. Olczak, A. M. Białkowska, and M. Koziołkiewicz, "Advanced Glycation End-Products (AGEs): Formation, Chemistry, Classification, Receptors, and Diseases Related to AGEs," Cells, vol. 11, no. 8. MDPI, Apr. 01, 2022. doi: 10.3390/cells11081312.
5. M. Gorska-Ciebiada, M. Saryusz-Wolska, A. Borkowska, M. Ciebiada, and J. Loba, "C-reactive protein, advanced glycation end products, and their receptor in type 2 diabetic, elderly patients with mild cognitive impairment," Front Aging Neurosci, vol. 7, no. OCT, 2015, doi: 10.3389/fnagi.2015.00209.
6. S. Sharif et al., "Low-grade inflammation as a risk factor for cardiovascular events and all-cause mortality in patients with type 2 diabetes," Cardiovasc Diabetol, vol. 20, no. 1, Dec. 2021, doi: 10.1186/s12933-021-01409-0.
7. J. Lee, J. S. Yun, and S. H. Ko, "Advanced Glycation End Products and Their Effect on Vascular Complications in Type 2 Diabetes Mellitus," Nutrients, vol. 14, no. 15. MDPI, Aug. 01, 2022. doi: 10.3390/nu14153086.
8. N. R. Sproston and J. J. Ashworth, "Role of C-reactive protein at sites of inflammation and infection," Frontiers in Immunology, vol. 9, no. APR. Frontiers Media S.A., Apr. 13, 2018. doi: 10.3389/fimmu.2018.00754.
9. D. Permatasari, B. Rachmawati, A. Riansari, E. Kurnia, and S. Limijadi, "Hubungan HbA1c dengan CRP pada Penderita Diabetes Melitus Tipe-2 Dengan Obesitas Dan Tanpa Obesitas," Journal of Nutrition College, vol. 9, pp. 267–272, 2020.
10. Y. Li et al., "Hs-CRP and all-cause, cardiovascular, and cancer mortality risk: A meta-analysis," Atherosclerosis, vol. 259, pp. 75–82, Apr. 2017, doi: 10.1016/j.atherosclerosis.2017.02.003.
11. L. Babu and A. Joshi, "Correlation of high-sensitivity C-reactive protein with blood sugar level in patients with Type 2 diabetes," Natl J Physiol Pharm Pharmacol, p. 1, 2017, doi: 10.5455/njppp.2018.8.0726805082017.

12. K. Basinska, K. Marycz, A. Śmieszek, and J. Nicpoń, "The production and distribution of IL-6 and TNF-α in subcutaneous adipose tissue and their correlation with serum concentrations in Welsh ponies with equine metabolic syndrome," J Vet Sci, vol. 16, no. 1, pp. 113–120, 2015, doi: 10.4142/jvs.2015.16.1.113.
13. V. P. Singh, A. Bali, N. Singh, and A. S. Jaggi, "Advanced glycation end products and diabetic complications," Korean Journal of Physiology and Pharmacology, vol. 18, no. 1. Korean Physiological Soc. and Korean Soc. of Pharmacology, pp. 1–14, 2014. doi: 10.4196/kjpp.2014.18.1.1.
14. S. Bansal, D. Chawla, M. Siddarth, B. D. Banerjee, S. V. Madhu, and A. K. Tripathi, "A study on serum advanced glycation end products and its association with oxidative stress and paraoxonase activity in type 2 diabetic patients with vascular complications," Clin Biochem, vol. 46, no. 1–2, pp. 109–114, Jan. 2013, doi: 10.1016/j.clinbiochem.2012.10.019.
15. A. A. Mostafa et al., "Plasma protein advanced glycation end products, carboxymethyl cysteine, and carboxyethyl cysteine, are elevated and related to nephropathy in patients with diabetes," Mol Cell Biochem, vol. 302, no. 1–2, pp. 35–42, Aug. 2007, doi: 10.1007/s11010-007-9422-9.
16. J. Lee, J. S. Yun, and S. H. Ko, "Advanced Glycation End Products and Their Effect on Vascular Complications in Type 2 Diabetes Mellitus," Nutrients, vol. 14, no. 15. MDPI, Aug. 01, 2022. doi: 10.3390/nu14153086.
17. K. Nowotny, T. Jung, A. Höhn, D. Weber, and T. Grune, "Advanced glycation end products and oxidative stress in type 2 diabetes mellitus," Biomolecules, vol. 5, no. 1. MDPI AG, pp. 194–222, Mar. 16, 2015. doi: 10.3390/biom5010194.
18. M. Gorska-Ciebiada, M. Saryusz-Wolska, A. Borkowska, M. Ciebiada, and J. Loba, "C-reactive protein, advanced glycation end products, and their receptor in type 2 diabetic, elderly patients with mild cognitive impairment," Front Aging Neurosci, vol. 7, no. OCT, 2015, doi: 10.3389/fnagi.2015.00209.
19. D. Indyk, A. Bronowicka-Szydełko, A. Gamian, and A. Kuzan, "Advanced glycation end products and their receptors in serum of patients with type 2 diabetes," Sci Rep, vol. 11, no. 1, Dec. 2021, doi: 10.1038/s41598-021-92630-0.
20. M. Khalid, G. Petroianu, and A. Adem, "Advanced Glycation End Products and Diabetes Mellitus: Mechanisms and Perspectives," Biomolecules, vol. 12, no. 4. MDPI, Apr. 01, 2022. doi: 10.3390/biom12040542.
21. V. P. Singh, A. Bali, N. Singh, and A. S. Jaggi, "Advanced glycation end products and diabetic complications," Korean Journal of Physiology and Pharmacology, vol. 18, no. 1. Korean Physiological Soc. and Korean Soc. of Pharmacology, pp. 1–14, 2014. doi: 10.4196/kjpp.2014.18.1.1.
22. J. Stanimirovic et al., "Role of C-Reactive Protein in Diabetic Inflammation," Mediators of Inflammation, vol. 2022. Hindawi Limited, 2022. doi: 10.1155/2022/3706508.
23. S. M. Nehring, A. Goyal, and B. C. Patel, "C reactive protein.," StatPearls Publishing, 2022.
24. J. F. Navarro et al., "Tumor necrosis factor-a gene expression in diabetic nephropathy: Relationship with urinary albumin excretion and effect of angiotensin-converting enzyme inhibition," 2005. [Online]. Available: http://frodo.wi.mit.edu/cgi-bin/primer3/
25. C. M. O. Volpe, P. H. Villar-Delfino, P. M. F. dos Anjos, and J. A. Nogueira-Machado, "Cellular death, reactive oxygen species (ROS) and diabetic complications review-Article," Cell Death Dis, vol. 9, no. 2, Feb. 2018, doi: 10.1038/s41419-017-0135-z.
26. Z. Aryan, A. Ghajar, S. Faghihi-Kashani, M. Afarideh, M. Nakhjavani, and A. Esteghamati, "Baseline High-Sensitivity C-Reactive Protein Predicts Macrovascular and Microvascular Complications of Type 2 Diabetes: A Population-Based Study," Ann Nutr Metab, vol. 72, no. 4, pp. 287–295, May 2018, doi: 10.1159/000488537.
27. M. Muniroh et al., "Effect of garcinia mangostana pericarp extract on glial nf-κb levels and expression of serum inflammation markers in an obese-type 2 diabetes mellitus animal model," Biomed Rep, vol. 15, no. 1, 2021, doi: 10.3892/br.2021.1439.
28. V. Cepas, M. Collino, J. C. Mayo, and R. M. Sainz, "Redox signaling and advanced glycation endproducts (AGEs) in diet-related diseases," Antioxidants, vol. 9, no. 2. MDPI, Feb. 01, 2020. doi: 10.3390/antiox9020142.
29. M. Pia de la Maza et al., "Fluorescent advanced glycation end-products (ages) detected by spectro-photofluorimetry, as a screening tool to detect diabetic microvascular complications," J Diabetes Mellitus, vol. 02, no. 02, pp. 221–226, 2012, doi: 10.4236/jdm.2012.22035.
30. S. H. Md Isa et al., "Improvement in C-reactive protein and advanced glycosylation end-products in poorly controlled diabetics is independent of glucose control," Diabetes Res Clin Pract, vol. 72, no. 1, pp. 48–52, Apr. 2006, doi: 10.1016/j.diabres.2005.09.011.
31. K. C. B. Tan, W.-S. Chow, S. Tam, R. Bucala, and J. Betteridge, "Association Between Acute-Phase Reactants and Advanced Glycation End Products in Type 2 Diabetes," 2004. [Online]. Available: http://diabetesjournals.org/care/article-pdf/27/1/223/563780/zdc00104000223.pdf
32. L. Shi, G. S. Tan, and K. Zhang, "Relationship of the Serum CRP Level With the Efficacy of Metformin in the Treatment of Type 2 Diabetes Mellitus: A Meta-Analysis," J Clin Lab Anal, vol. 30, no. 1, pp. 13–22, Jan. 2016, doi: 10.1002/jcla.21803.
33. P. Dwipayanan, I. Sarawati, and K. Suastika, "Perbandingan Kadar C-Reactive Protein Pada Penderita Diabetes Melitus Tipe 2 Yang Diterapi Dengan Insulin dan Obat Hipoglikemik Oral di Poliklinik Penyakit Dalam Rumah Sakit Umum Pusat Sanglah Denpasar Bali," Jurnal Penyakit Dalam Udayana, vol. 1, pp. 45–51, 2017, [Online]. Available: www.jpdunud.org
34. Z. Zhou et al., "Metformin Inhibits Advanced Glycation End Products-Induced Inflammatory Response in Murine Macrophages Partly through AMPK Activation and RAGE/NF B Pathway Suppression," J Diabetes Res, vol. 2016, 2016, doi: 10.1155/2016/4847812.
35. O. Horakova et al., "Metformin acutely lowers blood glucose levels by inhibition of intestinal glucose transport," Sci Rep, vol. 9, no. 1, Dec. 2019, doi: 10.1038/s41598-019-42531-0.
36. I. B. Hirsch, "Effect Of Insulin Therapy on Nonglycemic Variables During Acute Illness," ENDOCRINE PRACTICE, vol. 10, pp. 63–69, 2004.
37. S. Gallus et al., "Effect of Tobacco Smoking Cessation on C-Reactive Protein Levels in A Cohort of Low-Dose Computed Tomography Screening Participants," Sci Rep, vol. 8, no. 1, Dec. 2018, doi: 10.1038/s41598-018-29867-9.

# Effect of fetal bovine serum concentration towards vero cells growth on culture in DMEM medium

ORIGINALITY REPORT

| 18% | 13% | 12% | 3% |
|---|---|---|---|
| SIMILARITY INDEX | INTERNET SOURCES | PUBLICATIONS | STUDENT PAPERS |

PRIMARY SOURCES

| | | |
|---|---|---|
| 1 | Submitted to University of Kentucky<br>Student Paper | 1% |
| 2 | repository.umj.ac.id<br>Internet Source | 1% |
| 3 | www.ijbamr.com<br>Internet Source | 1% |
| 4 | www2.mdpi.com<br>Internet Source | 1% |
| 5 | Misganaw Asmamaw Mengstie, Endeshaw Chekol Abebe, Awgichew Behaile Teklemariam, Anemut Tilahun Mulu et al. "Endogenous advanced glycation end products in the pathogenesis of chronic diabetic complications", Frontiers in Molecular Biosciences, 2022<br>Publication | 1% |
| 6 | www.umfcluj.ro<br>Internet Source | 1% |
| | sites.kowsarpub.com | |

| | | |
|---|---|---|
| 7 | Internet Source | 1% |
| 8 | docksci.com<br>Internet Source | 1% |
| 9 | Jianming Zhu, Ziming Wang, Chunyan Lv, Mengtian Li, Kaiyi Wang, Zhencheng Chen. "Advanced Glycation End Products and Health: A Systematic Review", Annals of Biomedical Engineering, 2024<br>Publication | 1% |
| 10 | Jian Chen, Yanli Bi, Lei Chen, Qi Zhang, Linhao Xu. "Tanshinone IIA exerts neuroprotective effects on hippocampus-dependent cognitive impairments in diabetic rats by attenuating ER stress-induced apoptosis", Biomedicine & Pharmacotherapy, 2018<br>Publication | 1% |
| 11 | www.ncbi.nlm.nih.gov<br>Internet Source | <1% |
| 12 | Rehab Werida, Mahmoud Kabel, Gamal Omran, Ahmed Shokry, Tarek Mostafa. "Comparative clinical study evaluating the effect of adding Vildagliptin versus Glimepiride to ongoing Metformin therapy on diabetic patients with symptomatic coronary artery disease", Diabetes Research and Clinical Practice, 2020<br>Publication | <1% |

| | | |
|---|---|---|
| 13 | Yunwei Li, Xiaoming Zhong, Guanchang Cheng, Cuihua Zhao, Lei Zhang, Yan Hong, Qilin Wan, Ruili He, Zhizhong Wang. "Hs-CRP and all-cause, cardiovascular, and cancer mortality risk: A meta-analysis", Atherosclerosis, 2017<br>Publication | <1 % |
| 14 | www.phcogj.com<br>Internet Source | <1 % |
| 15 | Perdana Samekto Tyasnugroho Suyoto, Bianda Aulia. "Low muscle mass and inflammation among patients with type 2 diabetes mellitus in Indonesia", Diabetology International, 2018<br>Publication | <1 % |
| 16 | Stefano Genovese, Giorgia De Berardis, Antonio Nicolucci, Edoardo Mannucci et al. "Effect of Pioglitazone Versus Metformin on Cardiovascular Risk Markers in Type 2 Diabetes", Advances in Therapy, 2013<br>Publication | <1 % |
| 17 | coek.info<br>Internet Source | <1 % |
| 18 | thieme-connect.de<br>Internet Source | <1 % |
| 19 | f1000research.com<br>Internet Source | <1 % |

| | | |
|---|---|---|
| 20 | ojs.ptbioch.edu.pl<br>Internet Source | <1 % |
| 21 | Hemei Li, Jinling Zhou, Sitong Liu, Xiuting Chen, Tiantian Qin, Guodong Huang, Peizhen Luo, Ying Hu, Xing Xia. "Cinnamomum cassia Presl flavonoids prevent hyperglycemia-induced cognitive impairment via inhibiting of AGEs accumulation and oxidative stress", Journal of Functional Foods, 2023<br>Publication | <1 % |
| 22 | respiratory-research.biomedcentral.com<br>Internet Source | <1 % |
| 23 | www.spandidos-publications.com<br>Internet Source | <1 % |
| 24 | Hongdian Li, Mingxuan Li, Shaoning Dong, Ao Dong, Jing Wang, Yuanyuan Zhu, Yuanyuan Deng, Shu Chen, Mianzhi Zhang. "Preliminary study of the interactive effects of coronary heart disease and lacunar infarction on renal function in patients with type 2 diabetes mellitus by gender", Journal of Diabetes and its Complications, 2023<br>Publication | <1 % |
| 25 | Submitted to Universitas Negeri Surabaya<br>Student Paper | <1 % |
| 26 | mail.apicareonline.com<br>Internet Source | <1 % |

| | | |
|---|---|---|
| 27 | sciendo.com<br>Internet Source | <1% |
| 28 | G. Schernthaner. "Pleiotropic effects of thiazolidinediones on traditional and non-traditional atherosclerotic risk factors", International Journal of Clinical Practice, 2009<br>Publication | <1% |
| 29 | Ye Yao, Kaining Han, Shiyuan Dong, Mingyong Zeng, Zunying Liu. "Antioxidant balance and accumulation of advanced glycation end products after the consumption of standard diets including Maillard reaction products from silver carp peptides", Food & Function, 2016<br>Publication | <1% |
| 30 | publish.kne-publishing.com<br>Internet Source | <1% |
| 31 | www.robolabo.etsit.upm.es<br>Internet Source | <1% |
| 32 | Emir Muzurović, Zoja Stanković, Zlata Kovačević, Benida Š. Škrijelj, Dimitri P. Mikhailidis. "Inflammatory Markers Associated With Diabetes Mellitus – Old and New Players", Current Pharmaceutical Design, 2021<br>Publication | <1% |

| | | |
|---|---|---|
| 33 | Kalousova, M.. "Advanced glycoxidation end products in chronic diseases-clinical chemistry and genetic background", Mutation Research - Fundamental and Molecular Mechanisms of Mutagenesis, 20051111<br>Publication | <1 % |
| 34 | www.iscientific.org<br>Internet Source | <1 % |
| 35 | Hery Winarsi, Erminawati, Gumintang Ratna Ramadhan, Elsya Meliyana Gumelar, Lavia Androviterra Sekar Kencana. "Cowpea sprouted milk rich in phenolic antioxidants, vitamin C, protein, and dietary fiber as an antidiabetic drink", AIP Publishing, 2023<br>Publication | <1 % |
| 36 | Margherita Borriello, Fabio Lauria, Ivana Sirangelo, Krasimira Aleksandrova, Antje Hebestreit, Alfonso Siani, Paola Russo. "Association between Urinary Advanced Glycation End Products and Subclinical Inflammation in Children and Adolescents: Results from the Italian I.Family Cohort", Nutrients, 2022<br>Publication | <1 % |
| 37 | Marinos Kosmopoulos, Dimitrios Drekolias, Phaedon D. Zavras, Christina Piperi, Athanasios G. Papavassiliou. "Impact of advanced glycation end products (AGEs) | <1 % |

signaling in coronary artery disease", Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease, 2019
Publication

| | | |
|---|---|---|
| 38 | d-nb.info<br>Internet Source | <1 % |
| 39 | indonesianjournalofclinicalpathology.org<br>Internet Source | <1 % |
| 40 | jneuroinflammation.biomedcentral.com<br>Internet Source | <1 % |
| 41 | odr.chalmers.se<br>Internet Source | <1 % |
| 42 | www.biomed.bas.bg<br>Internet Source | <1 % |
| 43 | www.cadth.ca<br>Internet Source | <1 % |
| 44 | www.ejmanager.com<br>Internet Source | <1 % |
| 45 | www.med-sovet.pro<br>Internet Source | <1 % |
| 46 | www.tandfonline.com<br>Internet Source | <1 % |
| 47 | "OS01: Lung Cancer 1 : Oral Sessions", Respirology, 2013.<br>Publication | <1 % |

| | | |
|---|---|---|
| 48 | G. Schernthaner. "Pleiotropic effects of thiazolidinediones on traditional and non-traditional atherosclerotic risk factors", International Journal of Clinical Practice, 06/2009<br>Publication | <1 % |
| 49 | Asma Akhter, Sultan Alouffi, Uzma Shahab, Rihab Akasha et al. "Vitamin D supplementation modulates glycated hemoglobin (HBA1c) in diabetes mellitus", Archives of Biochemistry and Biophysics, 2024<br>Publication | <1 % |
| 50 | Clara Luna-Marco, Francesca Iannantuoni, Alberto Hermo-Argibay, Deédeni Devos et al. "Cardiovascular benefits of SGLT2 inhibitors and GLP-1 receptor agonists through effects on mitochondrial function and oxidative stress", Free Radical Biology and Medicine, 2024<br>Publication | <1 % |
| 51 | Oliver Lenz, Alessia Fornoni, Adeel Ijaz, Thor Tejada. "Role of Inflammation in Diabetic Nephropathy", Current Diabetes Reviews, 2008<br>Publication | <1 % |
| 52 | Takafumi Yoshida, Sho-ichi Yamagishi, Kazuo Nakamura, Takanori Matsui et al. "Pigment epithelium-derived factor (PEDF) inhibits | <1 % |

advanced glycation end product (AGE)-induced C-reactive protein expression in hepatoma cells by suppressing Rac-1 activation", FEBS Letters, 2006

Publication

| | | | |
|---|---|---|---|
| Exclude quotes | Off | Exclude matches | Off |
| Exclude bibliography | On | | |