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ORAL GLUCOSE TOLERANCE, HEPATOPROTECTIVE AND NEPHROPROTECTIVE EFFECT OF COMBINATION THERAPY OF LINAGLIPTIN AND *EMBLICA OFFICINALIS* AND ITS AMELIORATING EFFECT ON STREPTOZOTOCIN INDUCED DIABETIC RATS

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Keywords:

Aqueous fruit extract, Combination therapy, Oral glucose tolerance, Liver function indices, Kidney function indices

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ABSTRACT: Background & Objectives: *Emblica officinalis* Gaertn is a widely used medicinal plant with antidiabetic activity. This study was aimed to improve oral glucose tolerance, hepatoprotective and nephroprotective effect of linagliptin by given combination of aqueous fruit extract of *Emblica officinalis* on streptozotocin induced diabetic rats. **Materials & Methods:** Diabetes was produced on Wister albino rats by the intraperitoneal administration of streptozotocin (45 mg/kg BW). Linagliptin (5mg/70kg BW), *E. officinalis (*200 mg/kg BW) and fixed dose combination therapy of linagliptin (2.5 mg /70kg BW) with *E. officinalis* (100 mg/kg BW) were administered orally once daily for four weeks. **Results:** The result showed that both linagliptin and aqueous fruit extract of *E. officinalis* significantly increased oral glucose tolerance but combination therapy increased glucose utilization more significantly in comparison with linagliptin in glucose induced rats. Treatment with linagliptin and *E. officinalis* significantly restored liver function indices as measured by liver to body weight ratio, serum alanin transaminase, serum aspartate transaminase, serum alkaline phosphatase, serum Ɣ- glutamyl transferase, albumin and globulin and also kidney function indices as measured by kidney to body weight ratio, calcium, creatinine, urea and uric acid to normal levels. Where, their combination therapy significantly increased the hepatoprotective and nephroprotective activity in comparison with linagliptin. **Conclusion:** These investigations may suggest that combination therapy of linagliptin and aqueous fruit extract of *Emblica officinalis* might give higher ameliorating effect on oral glucose tolerance, hepatoprotective and nephroprotective activity in comparison with Linagliptin on streptozotocin induced diabetic rats.

INTRODUCTION: Today, diabetes is a serious concern for worldwide public health and its incidence is increasing daily. According to recent statistics, diabetes affects 4% of the world's population overall.

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This situation is quite concerning because by 2025, that number will have increased to 5.4%. This condition can rank as the 7th major cause of death by the year 2030.

Diabetes is a chronic condition that results in long term tissue damage and complications such dysfunctions of the liver and kidneys, which are usually connected to serious illness like organ failure [1]. Diabetes mellitus is associated with a marked increase in the risk of coronary heart disease (CHD) or stroke and cardiovascular disease (CVD), which is responsible for the majority of

deaths among patients with diabetes [2]. Plants are being thoroughly investigated for their potential to combat diabetes because of their immense therapeutic relevance. Herbal medications are often acceptable as they are recognized to have fewer side effects [3].

Emblica officinalis Gaertn, commonly known amloki in Bangladesh, is a member of the small genus of *Emblica* (Euphorbiaceae). It contents phenolic compounds and is a marked nutritional source of vitamin C, amino acid and minerals [4]. The aqueous fruit extract of *E. officinalis* has undergone pharmacological testing during the last ten years for a number of different purposes. These include antioxidant properties [5], also it is a powerful inhibitor of the generation of lipid peroxide and a scavenger of hydroxyl and superoxide radicals *in-vitro* due to it has antidiabetic activity [6].

Now, various types of drugs are used to treat diabetes. Only drugs are not enough to treat diabetes with CVD and other comorbities. Various medicinal plants have been used to the treatment of diabetes in combination with conventional drug [7]. Linagliptin is a type 2 diabetic medicine that is more affordable, well tolerated and safer. It works as an antidiabetic agent by lowering DPP4 activity thus controls the circulating levels of GIP and GLP-1 following meals. It declines the blood glucose levels without producing significant hypoglycaemia, the release of insulin is increased by GLP-1 and GIP. The GLP-1 also prevents glucagon secretion, which declines the post-meal rises in blood glucose and decreases fasting glucose levels [8]. Therefore, the current research is undertaken to evaluate the possible protective effect of linagliptin, *E. officinalis* and their combination therapy on streptozotocin induced diabetic rats. The purposes of this study was to investigate a safer, synergistic and promising hypoglycemic properties, cost effective, less toxic combination therapy by reducing dose level of oral hypoglycemic agents, while giving better glycemic control along with hepatoprotective and nephroprotective activity.

MATERIALS AND METHOD:

Raw Materials Collection: Streptozotocin used in this study was sourced from Sigma-Aldrich Chemical Company, Saint Louis, Missouri, USA. The rest of the chemicals employed were of standard analytical grade. The antidiabetic drug, linagliptin was collected from Square Pharmaceuticals Ltd, Bangladesh.

The Process for Making *Emblica officinalis* Aqueous Fruit Extract: Identification & Authentication of the *Emblica officinalis* was done in Botany Department, University of Rajshahi. The fruits were cleaned, dried in soft sunlight, and thus processed to a fine powder by using a grinder machine. In order to make the aqueous fruit extract, 500 g of the dried *E. officinalis* fruits were crushed in an electrical grinder and soaked in 2.5 L distilled water [9]. A magnetic stirrer was used to stir up the mixture for 24 hours at room temperature. After 24 hours later, with a fine sieve, the mixture was separated, and the crude extract was allowed to air-evaporate for three days [10]. The extract was then orally administered to the rats.

Selection of Animal: 25 Wister male rats weighing between 150 - 200 g (75 - 90 days old) were bought from the Pharmacy Department at Jahangirnagar University's Pharmacology Research Laboratory. All of the rats were acclimated to the new environmental conditions for one week prior to the experiments. The rats were housed in an animal facility with good ventilation and kept in a temperature-controlled environment of 25°C while receiving water and standard ICDDR, B pellets. All the rats were kept in cages that had a 12-hour cycle of natural light and dark. The institutional ethical committee of Varendra University in Bangladesh provided the ethical clearances. Randomly chosen Wister rats were placed in the normal control group (Normal, n = 5), Diabetic Control group (STZIDRs, n = 5), Diabetic group treating with 1 mL of 5 mg / 70kg BW of linagliptin (STZ + Linagliptin, n = 5), diabetic group treating with 1mL of 200 mg /kg BW aqueous fruit extract of *Emblica officinalis* (STZ + *Emblica officinalis,* n =5) and diabetic group treating with combination therapy of 1mL of 2.5 mg/70kg BW linagliptin and 100 mg/kg BW of aqueous fruit extract of *Emblica officinalis* (STZ+ combination, n = 5).

Experimental Induction of Diabetes: Two weeks of adaptation were given prior to the onset of diabetes. Except for the rats of normal control, all

other rats were not fed 16 h before injection and were given an intraperitoneal injection to produce them diabetic with a recently made single dose of streptozotocin (45 mg/kg BW). The streptozotocin (STZ) solution was produced by soaking in 0.01 M citrate buffer which were freshly made and adjusted to pH 4.5. To limit the early mortality as insulin reserves are released from injured pancreatic islets, rats were given drinking water laced with sugar (15g/L) after taking an STZ injection for 48 hours. Three days later, diabetes was identified by measuring blood glucose level using a glucometer (Bioland g- 423S Test strip, Germany) using rat tail vein blood samples. Rats having blood glucose level over 11.1 mmol/L, considered as diabetes were chosen for the experiment.

Evaluation of Oral Glucose Tolerance: After four weeks, the rats in groups (Diabetic Control group, STZ + Linagliptin, STZ + *Emblica officinalis* and STZ+ combination) were provided glucose (2 g/kg body weight) 30 mins after administration of the sample [11]. For the quick detection of blood glucose level, blood samples were taken from the tail vein prior to glucose administration and at 30, 60, and 90 min after glucose administration.

Measurement of Liver and Kidney Function Indices: After a four-week experimental period, every single rat liver to body weight ratio and kidney to body weight ratio were calculated. The serum concentrations of creatinine, [12] urea, [13] calcium, [14] uric acid, [15] albumin, and globulin [16] as well as the activities of serum alkaline phosphatase (ALP), [17] serum gamma glutamyl transferase (GGT) [18] using Randox assay kits and serum aspartate transferase (AST) [19] and serum alanine transaminase (ALT) [19] were determined in the serum by taking UV absorbance, using diagnostic kits (Human Germany).

Statistical Analysis: Using Microsoft Office excel 2007, the results were reported as mean ± SEM and IBM SPSS statistics 23. We used a one-way analysis of variance (ANOVA), followed by Dunnet's test. The statistical method was applied on each analysis was described in each figure and table. Result was taken as significant when P values were less than 0.01(P<0.01).

RESULTS: The oral glucose tolerance, liver and kidney function activity were measured as part of a four-week protocol to observe the results of linagliptin, *E. officinalis* and its combination on streptozotocin induced diabetic rats. Linagliptin was used as standard antidiabetic agents and *E. officinalis* as an antidiabetic medicinal plant.

Effect of Linagliptin, Aqueous Fruit Extract of *Emblica officinalis* and their Combination Therapy on Oral Glucose Tolerance:

TABLE 1: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN, *EMBLICA OFFICINALIS* AND THEIR COMBINATION THERAPY ON ORAL GLUCOSE TOLERANCE IN STREPTOZOTOCIN INDUCED DIABETIC RATS: (N= 5, MEAN ±SEM)

Group	Fasting	30 minutes	60 minutes	90 minutes
Normal	5.52 ± 0.35	5.78 ± 0.56	4.89 ± 0.28	4.52 ± 0.59
STZIDRs	23.06 ± .99#	32.64 ± 1.08 #	29.85 ± 1.02 #	24.25 ± 1.21 #
STZ+ Linagliptin	7.25 ± 0.98 *	16.48 ± 1.03	13.58 ± 0.89	10.56 ± 0.89 *
STZ+ *Emblica officinalis*	6.82 ± 0.79 *	15.77 ± 0.95	12.55 ± 0.86	9.56 ± 1.11 *
STZ+ combination	6.12 ± 0.98 **	13.46 ± 0.87	9.56 ± 0.58 *	6.42 ± 0.45 **

Where significant value is *p<0.05 compared to normal group and **p<0.01 compared to diabetic control group (ANOVA followed by Dunnett's test). Here, STZ + Linagliptin, (5mg/70kg BW), STZ + *Emblica officinalis* (200 mg/kg BW) and STZ+ combination (linagliptin (2.5 mg /70kg BW) with E. officinalis (100 mg/kg BW)).

Table 1 observes the blood glucose levels of rats after orally administration of glucose. In diabetic control group (STZIDRs) blood glucose level raised to the peak at 30 min after glucose taken and next 60 mins it stayed high. Linagliptin, *E. officinalis* and their combination therapy observed significant decrease (*P< 0.05 and **P< 0.01) in blood glucose concentration at 60 and 90 min compared with diabetic control rats. Here, combination therapy showed higher oral glucose tolerances in comparison with Linagliptin.

Effect of Linagliptin, Aqueous Fruit Extract of *Emblica officinalis* and their Combination

Therapy on Parameters for Liver Function: The diabetic control groups (STZIDRs) showed significant higher level (*P< 0.05) of liver to body weight ratio, serum of alanine transaminase (ALT), aspartate transaminase (ALT), alkaline phosphatase (ALP), gama glutamyl transferase (GGT) and globulin, as well as significant lower level of albumin concentration when compared with the normal control group in the **Fig. 1, 2, 3** and **4**. continuous administration of linagliptin, E. *officinalis* and their combination therapy to diabetic rats for four weeks was able to restore all the liver function indices back to normalcy. The combination therapy significantly decreased (**P<0.01) level of the liver to body weight ratio 37.53%, serum ALT 44.21%, AST 36.89 %, ALP 57.49%, GGT 51.84%, Globulin 13.06 % level and increased the Albumin 17.26% in comparison with Linagliptin.

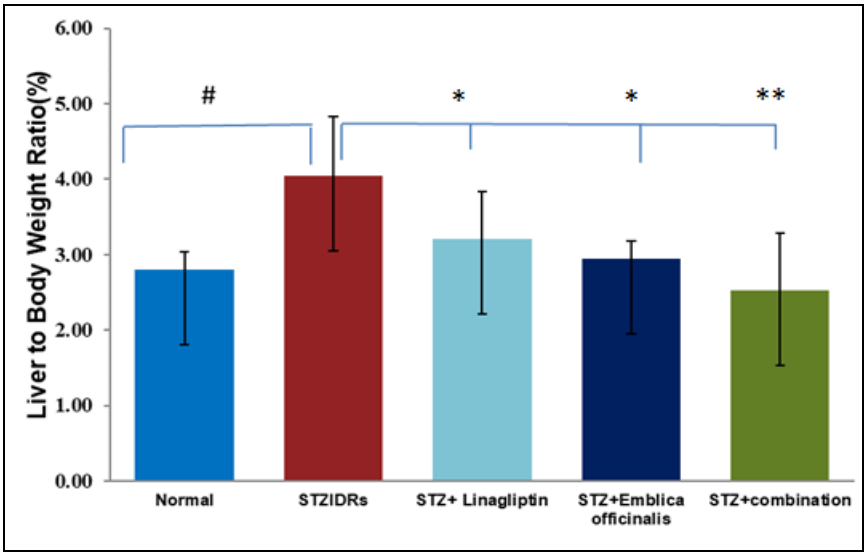

FIG. 1: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN (5 mg/70 kg BW), *EMBLICA OFFICINALIS* (200 mg/ kg BW) AND THEIR COMBINATION THERAPY (LINAGLIPTIN (2.5 mg /70 kg BW) WITH *E. OFFICINALIS* (100 mg/kg BW) ON LIVER TO BODY WEIGHT RATIO IN STREPTOZOTOCIN INDUCED DIABETIC RATS (STZIDRS): (n= 5, mean ±SEM). ANOVA followed by Dunnett's test. Where significant value is *p< 0.05 and **p<0.01 when compared to diabetic control group #: significantly different *p<0.05 from normal control group.

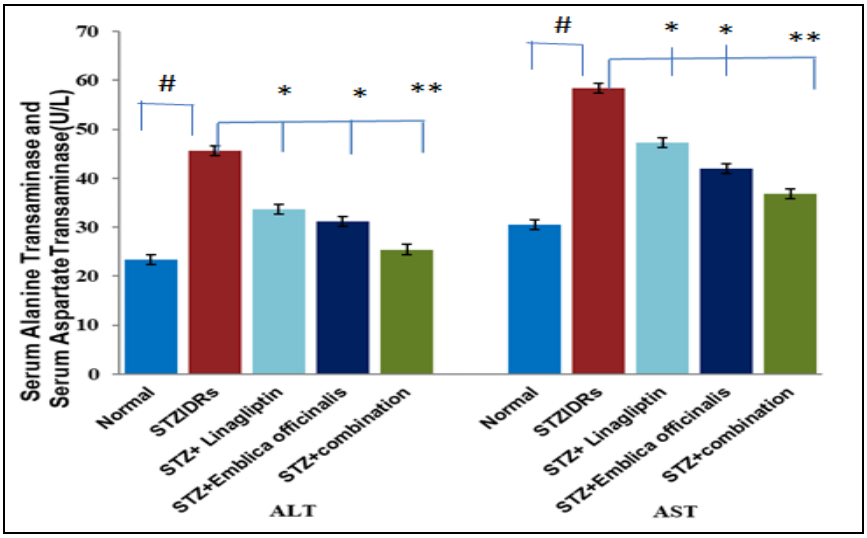

FIG. 2: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN (5 mg/70 kg BW), *EMBLICA OFFICINALIS* (200 mg/ kg BW) AND THEIR COMBINATION THERAPY (LINAGLIPTIN (2.5 mg /70 kg BW) WITH *E. OFFICINALIS* (100 mg/kg BW) ON SERUM ALANINE TRANSAMINASE (ALT) AND SERUM ASPARTATE TRANSFERASE (AST) IN STREPTOZOTOCIN INDUCED DIABETIC RATS (STZIDRS): (n= 5, mean ±SEM). ANOVA followed by Dunnett's test. Where significant value is *p<0.05 and **p<0.01 when compared to diabetic control group #: significantly different *p<0.05 from normal control group.

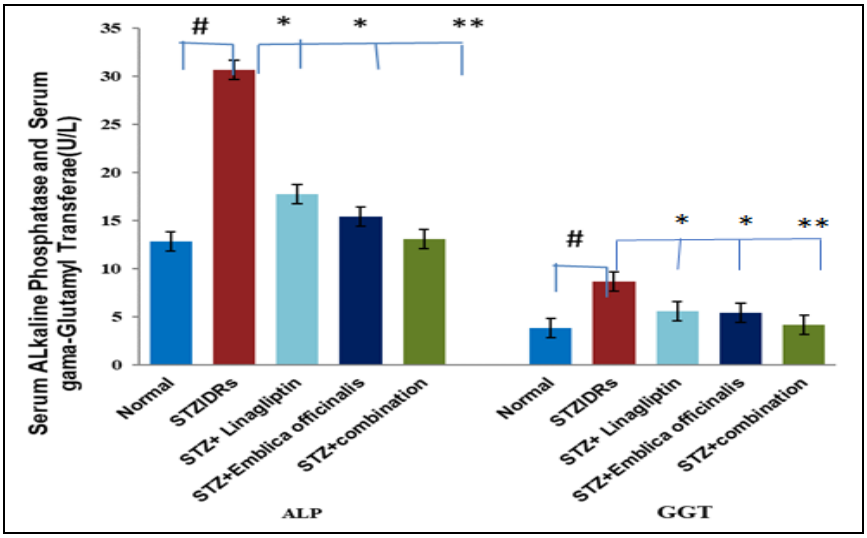

FIG. 3: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN (5 mg/70 kg BW), *EMBLICA OFFICINALIS* (200 mg/ kg BW) AND THEIR COMBINATION THERAPY (LINAGLIPTIN (2.5 mg /70 kg BW) WITH *E. OFFICINALIS* (100 mg/kg BW) ON SERUM ALKALINE PHOSPHATASE (ALP) AND SERUM GAMA GLUTAMYL TRANSFERASE (GGT) IN STREPTOZOTOCIN INDUCED DIABETIC RATS (STZIDRS): (n= 5, mean ±SEM). ANOVA followed by Dunnett's test. Where significant value is *p<0.05 and **p<0.01 when compared to diabetic control group #: significantly different *p<0.05 from normal control group.

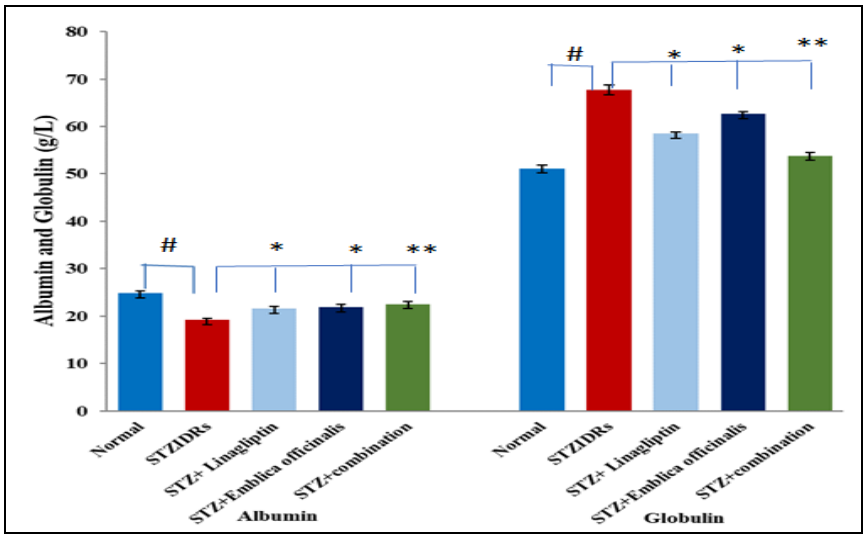

FIG. 4: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN (5 mg/70 kg BW), *EMBLICA OFFICINALIS* (200 mg/ kg BW) AND THEIR COMBINATION THERAPY (LINAGLIPTIN (2.5 mg /70kg BW) WITH *E. OFFICINALIS* (100 mg/kg BW) ON SERUM ALBUMIN AND GLOBULIN IN STREPTOZOTOCIN INDUCED DIABETIC RATS (STZIDRS): (n= 5, mean ± SEM). ANOVA followed by Dunnett's test. Where significant value is *p<0.05 and **p< 0.01 when compared to diabetic control group #: significantly different *p<0.05 from normal control group.

Effect of Linagliptin, Aqueous Fruit Extract of *Emblica officinalis* and their Combination Therapy on Parameters for Kidney Function:

TABLE 2: EFFECT OF FOUR WEEKS TREATMENT WITH LINAGLIPTIN, *EMBLICA OFFICINALIS* AND THEIR COMBINATION THERAPY ON THE PARAMETERS OF KIDNEY FUNCTION IN STREPTOZOTOCIN INDUCED DIABETIC RATS: (N= 5, MEAN ±SEM)

Parameters of kidney function	Normal	STZIDRs	STZ+ Linagliptin	STZ+ *Emblica officinalis*	STZ+ combination
Kidney to body weight ratio (%)	5.28 ± 0.22	8.77 ± 0.53 #	6.95 ± 0.76*	6.25 ± 0.86*	5.72 ± 0.86 **
Calcium (mmol/L)	1.42 ± 0.054	3.25 ± 0.160 #	2.20± 0.129 *	2.03 ± 0.064*	1.87 ± 0.050 **
Creatinine (mg/dl)	0.634 ± 0.018	1.47 ± 0.034 #	0.976 ± 0.029 *	0.852 ± 0.018 *	0.774 ± 0.027 **

Urea (mg/dl)	34.9 ± 0.582	168.5 ± 1.361 #	88.6 ± 1.218*	75.9± 1.077 *	48.9 ± 0.910 **
Uric acid (mg/dl)	5.06 ± 0.255	9.39 ± 0.357 #	7.23 ± 0.409 *	7.13 ± 0.303 *	5.53 ± 0.196 **

Where significant value is *p<0.05 compared to normal group and **p<0.01 compared to diabetic control group (ANOVA followed by Dunnett's test). Here, STZ + Linagliptin, (5mg/70kg BW), STZ + *Emblica officinalis* (200 mg/kg BW) and STZ+ combination (linagliptin (2.5 mg /70kg BW) with *E. officinalis* (100 mg/kg BW).

After four weeks the diabetic control groups (STZIDRs) showed significant increased (*P< 0.05) in kidney to body weight ratio and in serum concentration of calcium, creatinine, urea and uric acid when compared with the normal control group in the **Table 2**. On the other hand, continuous administration of linagliptin, *E. officinalis* and their combination therapy to diabetic rats for four weeks was able to restore all the kidney function indices back to normalcy. Here, The combination therapy significantly decreased (**P<0.01) level of the Kidney to body weight ratio 34.78%, Serum Calcium 42.46%, Creatinine 47.35%, Urea 70.97% and Uric acid 41.09% in comparison with Linagliptin.

DISCUSSION: In this study linagliptin, *E. officinalis* and their combination therapy increased glucose utilization in glucose induced rat. The improving liver to body weight ratio in diabetic control group observed in the experiment may be an indication of liver inflammation [20]. It most likely lead to the increase in ALP [21] as reduce in albumin and globulin concentration. GGT is an essential enzyme for the metabolism of glutathione in the liver that is confined to membranes [22]. So, the rise level of serum ALP and GGT concentration in the diabetic control group leads the damage to the plasma membrane [23]. To predict probable liver injury, the well-known transaminase ALT and AST are used as markers [24].

The increased serum level of both transaminase, were observed in diabetic control rats, and showed liver cell injury as well [25]. Continuous oral administration of linagliptin, *E. officinalis* and their combination therapy to diabetic rats reduced the activities of ALT and AST enzyme comparable to the diabetic control group. Same results are shown utilizing *Eugenia jambolana* seed kernels in STZ induced diabetic rats, according to Ravi *et al* [26]. The condition of liver is determined by the levels of albumin and globulin (protein molecule). Globulins are bigger proteins that are in charge of immunologic reactions, albumin is produced in the liver, is a significant carrier protein which circulates in the bloodstream [27]. Low level of serum albumin and high level of globulin indicate to infection related chronic liver damage [28]. So, the decrease level of serum albumin and increase level of globulin in diabetic control rats is an indication of liver damage. Oral administration of linagliptin, *E. officinalis* and their combination therapy to diabetic rats restored the albumin and globulin levels to normalcy.

As observed in the kidney of untreated diabetic rats, the significant increase in kidney-to-body weight ratio may also be a result of inflammation [20]. The increased serum levels of calcium ion, creatinine, urea and uric acid are indicated of kidney injury. Diabetes are compelled to go through an osmotic diuretics, which leads to electrolyte loss and dehydration when glucose is excreted in urine. An attempt by the kidney to buffer the urine decreases electrolytes such as calcium in serum [29]. Oral administration of linagliptin, *E. officinalis* and their combination therapy to diabetic rats significantly reduced the serum calcium levels when compared with diabetic control group, the decline was more pronounced in the combination treatment.

So, it may help the kidney regain their ability to regulate osmotic pressure. The increase in serum levels of urea, creatinine, and uric acid in the untreated diabetic rats as observed in the present study. The inability of glucose to reach the extra hepatic tissues as a result of insulin insufficiency triggers gluconeogenesis, which acts as a source of glucose in place of insulin [5]. In order to support this route, increased proteolysis releases free glucogenic amino acids into the plasma. These amino acids are then deaminated in the liver, which raises serum urea levels. Oral administration of linagliptin, *E. officinalis* and their combination therapy to diabetic rats produced a significant reduction in the levels of these three metabolites,

consequently providing defense against diabetes-related dysfunction. Similar results were observed in diabetic rats given extracts from *Picrorhiza kurroa* and *Vernonia* [30].

CONCLUSION: The results observed from these study examined that the linagliptin and aqueous fruit extract of *Emblica officinalis* significantly increased oral glucose tolerance, hepatoprtective and nephroprotective activity. But oral administration of their combination therapy might give higher ameliorating effect on oral glucose tolerance, hepatoprotective and nephroprotective activity in comparison with their monotherapy alone on streptozotocin induced diabetic rats. As a result, this combination therapy may be an excellent, less harmful and cost-effective treatment for diabetes. Further study is necessary for the tolerability, pharmacodynamic, pharmacokinetic and drug action of this fixed dose combination therapy (linagliptin and *E. officinalis*) that might reveal new agent for the treatment of diabetes.

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CONFLICTS OF INTEREST: None

REFERENCES:

1. WHO Global status report on non communicable diseases. http://www.who.int/nmh/ publications/ ncd_report_full_en.pdf, 2012.
2. Masumoto A, Hirooka Y, Shimokawa H, Hironaga K and Setoguchi S. Takeshita A: Possible involvement of Rho-kinase in the pathogenesis of hypertension in humans. Hypertension 2001; 38: 1307-1310.
3. Modak M, Dixit P, Londhe J, Ghaskadbi S and Devasagayam TP: Indian herbs and herbal drugs used for the treatment of diabetes. J Clin Biochem Nutr 2007; 40:1 63–73.
4. Anila L and Vijayalakshmi NR: Flavonoids from *Emblica officinalis* and *Mangifera indica*, effectiveness for dyslipidemia. J Ethnopharmacol 2002; 79: 81–87.
5. Po-Hsien Li, Chien-Wen Wang, Wen-Chien Lu, Tuzz-Ying Song and Chiun-C. R. Wang: Antioxidant, Anti-Inflammatory Activities, and Neuroprotective Behaviors of *Phyllanthus emblica* L. Fruit Extracts. Agriculture 2022; 12(5): 588.
6. Anterpreet K. Chahal, Gourav Chandan, Rakesh Kumar, Anil Kumar Chhillar, Adesh K. Saini and Reena V. Saini: Bioactive constituents of *Emblica officinalis* overcome oxidative stress in mammalian cells by inhibiting hyperoxidation of peroxiredoxins. J of Food Biochem 2020; 44(2): 13115.
7. Malviya N, Jain S and Malviya S: Antidiabetic potential of medicinal plants. Acta Pol Pharm Drug Res 2010; 67: 113-18.
8. Drucker D, Easley C and Kirkpatrick P: News and analysis. Nature Reviews Drug Discovery 2007; 6: 109-10.
9. Gohil T, PathakN, N Jivani, Devmurari V and Patel J: Treatment with extracts of *Eugenia jambolana* seed and *Aegle marmelos* leaf extracts prevents hyperglycemia and hyperlipidemia in alloxan induced diabetic rats. Afr J Pharm Pharmacol 2010; 4: 270-275.
10. El Amin M, Virk P, Elobeid M, Almarhoon Z, Hassan Z, Omer S, Merghani N, Daghestani M and Al Olayan E: Anti-diabetic effect of *Murraya koenigii* (L) and *Olea europaea* (L) leaf extracts on streptozotocin induced diabetic rats. Pak J Pharm Sci 2013; 26: 359-365.
11. Joy KL and Kuttan R: Anti-diabetic activity of *Picrorrhiza kurroa* extract. J of Ethnopharma 2003; 67(2): 143–48.
12. Toora BD: Measurement of creatinine by Jaffe's reaction--determination of concentration of sodium hydroxide required for maximum color development in standard, urine and protein free filtrate of serum 2002; 40(3): 352-4.
13. Marsh WH, Fingerhut B and Miller H: Automated and manual direct methods for the determination of blood urea. Clinical Chemistry 1965: 11: 624–627.
14. Tietz NW, Prude EL and Sirgard-Anderson O: Tietz Text-book of Clinical Chemistry, WB Saunders Company London UK 1994.
15. Tietz NW: Clinical Guide to laboratory Tests, WB Saunders, Philadelphia, Pa, USA, 3rd Edition 1995.
16. Spencer K and Price CP. Influence of reagent quality and reaction conditions on the determination of serum albumin by the bromcresol green dye binding method. Annals of Clinical Biochemistry 1977; 14(2): 105–115.
17. Wright PJ, Leathwood PD and Plummer DT: Enzymes in rat urine: alkaline phosphatase. Enzymologia 1972; 42(4): 317–327.
18. Szasz G: A kinetic photometric method for serum gamma-glutamyl transpeptidase. Clinical Chemistry 1996; 15(2): 124–136.
19. Pulye Roy, Monirul Islam, Mohammad Saiful Islam and Mamunur Rashid: Beneficial Effects of Combination Therapy of Sitagliptin and β-carotene Drugs on Streptozotocin-induced Diabetic Rats. Bangladesh Pharmaceutical Journal 2020; 23(2): 87-95.
20. Moore KL and Dalley AF: Clinical Oriented Anatomy, Woller Klumner Corporation Philadelphia 1999.
21. Akanji MA: Effect of chronic consumption of metabisulphite on the integrity of the rat kidney cellular system. Toxicology 1993; 81(3): 173–179.
22. Kaplan LA and Pesce AJ: Clinical Chemistry: Theory, Analysis and Correlation, Harcourt Health Science Company London UK 1996.
23. Yakubu MT, Bilbis LS, Lawal M and Akanji MA: Effect of repeated administration of sildenafil citrate on selected enzyme activities of liver and kidney of male albino rats. Nigerian J of Pure Appl Science 2003; 18(1): 1395–1400.
24. Rahman MF. Effects of vepacide (*Azadirachta indica*) on aspartate and alanine aminotransferase profiles in a subchronic study with rats. Human and Experimental Toxicology 2001; 20(5): 243–249.
25. Wolf PL, Williams D, Tsudaka T and Acosta L: Methods and Techniques in Clinical Chemistry John Wiley & Sons, New York NY USA 1972.
26. Ravi K, Sivagnanam K and Subramanian S: Anti-diabetic activity of *Eugenia jambolana* seed kernels on

streptozotocin- induced diabetic rats. Journal of Medicinal Food 2004; 7(2): 187–191.
27. Tietz NW: Fundamentals of Clinical Chemistry WB Saunders Philadelphia Pa USA 1986.
28. Naganna B, Talwar GP, Srivastava LM and Moudgils KD. Plasma proteinseds in Textbook of Biochemistry and Human Biology. Eds 1999; 172–180.
29. Ramsey IA: A Synopsis of Endocrinology and Metabolism. Wright Publisher Bristol UK 1986.
30. Mohammed Rahmatullah, Taslima Ferdousi Shefa, Labiba Hasan, Md. Tozammal Hossain, Salman Ahmed, Abdullah Al Mamun, Md. Rasadul Islam, Shahnaz Rahman and Majeedul H. Chowdhury: Study on Antinociceptive and Anti-hyperglycemic Activity of Methanol Extract of *Brassica juncea* (L.) Czern. Leaves in Mice. Advances in Natural and Applied Sciences 2010; 4(3): 221-225.

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