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EVALUATION OF ANTI-OBESITY & HYPOLIPIDEMIC ACTIVITIES OF ETHANOLIC EXTRACT OF *CINNAMOMUM ZEYLANICUM* AND *MORUS ALBA*

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ABSTRACT

It refers the evaluation of anti-obesity and hypolipidemic activities of ethanolic extract of *Cinnamomum zeylanicum* and *Morus alba.* The fresh bark of *Cinnamomum zeylanicum* and leaves of *Morus alba* were collected from UP East region and authenticated from a botanist. Bark of *Cinnamomum zeylanicum* was extracted through Soxhlet and leaves of *Morus alba* extracted through maceration process using ethanol. The albino rats were obtained from the Animal House, Saraswati Higher Education and Technical College of Pharmacy, Gahni, Ayar, Varanasi; kept at temp. 25±1°C with 12-hour light/dark cycle. Animals were kept into 5 groups; n=6 i.e., group 1: normal saline; group 2: high fat diet (HFD) + normal saline; group 3: HFD + *Cinnamomum zeylanicum* (400mg/kg); group 4: HFD + *Morus alba* (400mg/kg) for 15 days and group 5 given HFD + Orlistat (50mg/kg) for 15 days. The blood sugar level was estimated at the day 1, 3, 5, 10 & 15. The HFD + *Cinnamomum zeylanicum* (400mg/kg) treated animals showed the blood glucose level as 90.43±0.42mg/dl, 92.20±0.23mg/dl and 94.20±0.34mg/dl, at the day 5, day 10 and day 15, respectively. However, HFD + *Morus alba* (400mg/kg) treated rats showed the blood glucose level as 91.45±0.64mg/dl, 94.46±0.25mg/dl and 96.20±0.12mg/dl, respectively. This study suggests that *Cinnamomum zeylanicum* and *Morus alba* medication may be recommended as a well

recognised treatment for obesity management and cholesterol reduction, thereby improving overall health and extending lifespan.

KEYWORDS: *Cinnamomum zeylanicum, Morus alba,* anti-obesity, hypolipidemic, orlistat.

INTRODUCTION

Obesity is a term used to describe a medical condition that is sometimes used to describe excessive body fat storage that is harmful to human health. In 2016, it was discovered that over 1.9 billion adults were fat. 2020 saw the examination of almost 39 million youngsters (≤5 years) for overweight (WHO, 2021). Treatment is dependent on cause and condition's severity that includes lifestyle changes, heart-healthy foods and increased physical activity. It also uses medicines for weight loss approved by FDA. For some people, surgical procedure may be also used (Safaei et al. 2021). Obesity is a worldwide issue that affects an estimated 300 million individuals. In the three decades between 1980 and 2008, rates of obesity increased worldwide (Finucane et al. 2011). Although the prevalence of obesity is higher in upper-middle-class and wealthy nations, it is predicted that it will climb swiftly in lower-income nations (Kelly et al. 2008).

Cinnamomum zeylanicum (CZ)

It is often referred to as "true cinnamon" or Ceylon cinnamon (from which its Latin name, zeylanicum) (Singh et al. 2007). Three major constituents comprising 82.5% of the total composition of the essential oils extracted from CZ bark(Simic et al. 2004). The main constituents of CZ extracts are also eugenol and cinnamonaldehyde (Usta et al. 2003). Because of its distinctive characteristics, C. zeylanicum can be easily distinguished from other species of cinnamon. It can reach a height of 12 meters and is typically grows in soil that is lateritic, loamy, and contains silver sand. The greenish-colored flowers of C. zeylanicum are grouped in panicles from either the axial or apical position (Azad et al. 2016). The distinct green color of C. zeylanicum blossoms distinguishes them from those of other Cinnamon types. (Ariyarathne et al. 2018).

Morus alba

For everyone's eternal health, longevity, and healing, to get rid of suffering and anxiety, and for food, flavor, and aroma, humans from all over the world are dependent on the plant kingdom to meet their demands. The species is a rapidly growing tree which cgrow up to 20 meters in height. The trees are trimmed to ease the gathering of leaves or fruits under

cultivation with regular harvesting. The bark is a dark grey-brown colour with lenticels running horizontally. The leaves are glossy green and cordate at the apex, with serrated margins and long petioles. The length ranges from 5.0-7.5cm (Devi et al. 2013).

They can be found in Asia's subtropical regions. They can also be found in other parts of the world i.e., North America, Africa. The first nine species documented so far are found in Asia, while the latter five are recently discovered. Mulberry fruits are often utilized as important foods because they contain various vital like linnolenic acid or oleic acid. These unsaturated fatty acids are essential for cell membrane synthesis, optimal brain function, hormone secretion and production, immunological function and inflammatory response management, and blood pressure modulation (Carrizzo et al. 2016). It's also used to make silkworms and ingredients for sericulture. Mulberry fruits like mulberry pekmex, mulberry Kestil, and mulberry Kome are most commonly found in Turkey. This fruit is used in Turkey to make medication, juice, natural colours, and industrial cosmetics (Zeng et al. 2015).

MATERIALS AND METHODS

Experimental requirements

Cinnamomum zeylanicum, *Morus alba*, High Fat Diet (HFD), distilled water, water-bath, electronic weight balance & ethanol.

Collection, authentication, and extraction of the plant

The fresh bark of *Cinnamomum zeylanicum* and leaves of *Morus alba* were collected from UP East region and authenticated from a botanist. The bark was cleaned to remove any dust and allowed to air dry or be covered with shade. The bark and leaves that had dried were ground into coarse and then fine powders. Weighed out powder was steeped in ethanol solvent for 15 days, stirring gradually during that time. A rotary evaporator was used to partially vacuum-dry the mixed slurry that was produced. The *Cinnamomum zeylanicum* and *Morus alba* extract's percentage yield was calculated thru below formula:

$$\text{percent yield} = \frac{\text{actual yield}}{\text{theoretical yield}} \times 100\%$$

Preparation of animals

From the Animal House, Saraswati Higher Education and Technical College of Pharmacy, Gahni, Ayar, Varanasi albino rats were acquired. The rats were kept in ideal conditions, with

a 12 hrs light/dark cycle and temp. 25±1°C. They were fed a regular mouse pellet diet and given access to unlimited water, with the RH being kept at 44-56%. Rats are denied food one hour before the experiment (Bhajoni, 2016).

Experimental design

Rats were kept in different 5 groups; n=6 and dosed for 15 days:

Group	Treatment
1	Normal saline
2	High fat diet
3	High fat diet + *Cinnamomum zeylanicum* (400mg/kg).
4	High fat diet + *Morus alba* (400mg/kg).
5	High fat diet + Orlistat (50mg/kg).

Evaluation parameters

Body weight

Before the medicine is administered and when the dosage is finished, the weight of every animal in each is measured. Body weight is compared before and after the medication is administered.

Weigh of organs

After their sacrifice, all of the rats from the various groups are mutilated. Organs including the brain, kidney, and lever were all weighed individually to confirm the effect on various organs.

Total cholesterol level

Total Cholesterol testing was carried out correctly and in accordance with guidelines. Every test was carried out utilizing the particular medical kits that were on the market. A medical kit that was purchased from a certified manufacturer and supplier was also used to assess the amount of triglycerides in blood plasma (Dholi et al. 2011).

Food consumption pattern

Rats' eating habits are examined on days 5, 10, and 15. An hour before to the experiment, the rats are fed a high-fat diet and are not allowed to eat. Groups of rats in their cages were fed 10 g of rat pellets after 30 minutes of progesterone administration, and food ingestion was monitored at 0.5, 1 & 1.5 hrs intervals (Nderitu et al. 2017).

Blood sugar level

Seven blood glucose readings are taken at 0,5,10, and 15 days following the initiation of medication therapy. Dr. Morepen's blood glucometer is used to estimate blood glucose levels after a blood sample is taken by cutting the tail. It is a simple and genuine process (Jung et al. 2021).

RESULTS AND DISCUSSION

Percentage yield

The fresh bark of *Cinnamomum zeylanicum* and leaves of *Morus alba* were demonstrated the percentage yield as 46.24% and 42.61%, respectively.

Evaluation of anti-obesity potential

Body weight measurements

The group given with normal saline showed a substantial rise in body weight, increasing from 130g initial weight to 142g post-treatment. The HFD resulted in a significant rise in body weight, reaching 140g compared to 175g before the therapeutic intervention.

After 15 days of exposure, HFD + *Cinnamomum zeylanicum* (400mg/kg) and HFD + *Morus alba* (400mg/kg) demonstrated the body weight as 155g and 160g. The HFD + Orlistat (50mg/kg) treated group exhibited marked decrease in body weight as 150g Thus, both the herbs-maintained body weights of rats. However, the effect of *Cinnamomum zeylanicum* was much significant that *Morus alba.*

Table 1: Body weight measurements after 15 days.

Treatment	Body weight (g)	
	Before treatment	After treatment
Normal saline	130g	142g
HFD	140g	175g
HFD + *Cinnamomum zeylanicum* (400mg/kg)	130g	155g
HFD + *Morus alba* (400mg/kg)	130g	160g
HFD + Orlistat (50mg/kg)	140g	150g

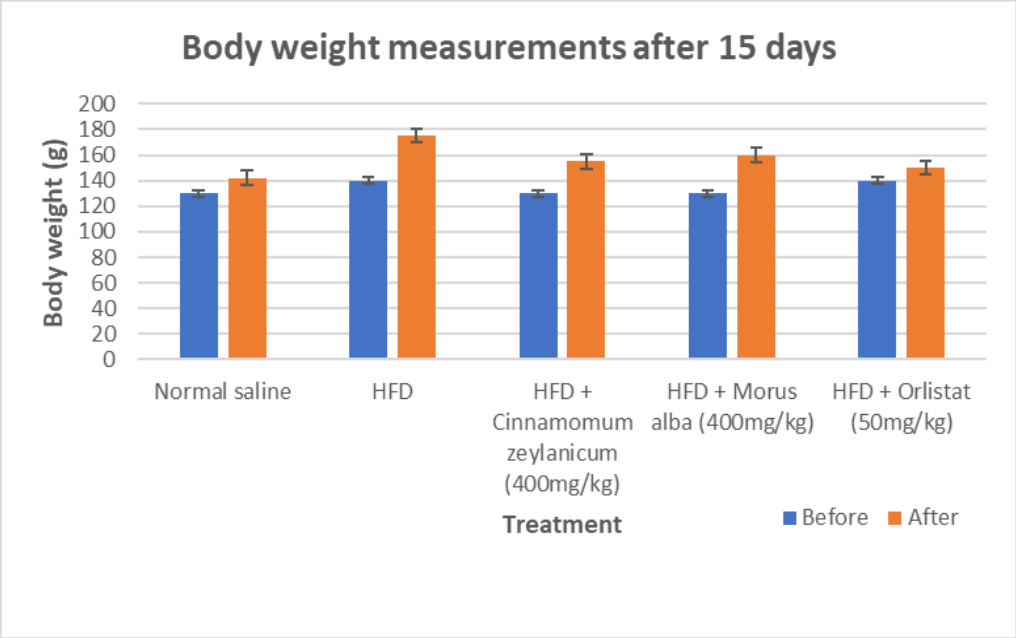

Fig. 1: Graphical data of body weight measurements after 15 days.

Weight of organs measurements

HFD + *Cinnamomum zeylanicum* (400mg/kg) and HFD + *Morus alba* (400mg/kg) treated rats showed a strong weights management of different organs i.e., spleen, kidney, and liver. The weight of spleen was observed as 0.59±0.35g, 0.57±0.24g in the HFD + *Cinnamomum zeylanicum* (400mg/kg) and HFD + *Morus alba* (400mg/kg) treated groups, respectively. Similar effect was also found on kidney and liver.

Table 2: Weight of organs measurements.

Treatment	Spleen (g)	Kidney (g)	Liver (g)
Normal saline	0.56±0.12	1.17±0.24	7.37±0.12
HFD	0.59±0.18	1.98±0.27	9.20±0.28
HFD + *Cinnamomum zeylanicum* (400mg/kg)	0.59±0.35	1.53±0.13	7.67±0.16
HFD + *Morus alba* (400mg/kg)	0.57±0.24	1.72±0.45	8.12±0.19
HFD + Orlistat (50mg/kg)	0.51±0.25	1.26±0.26	7.10±0.63

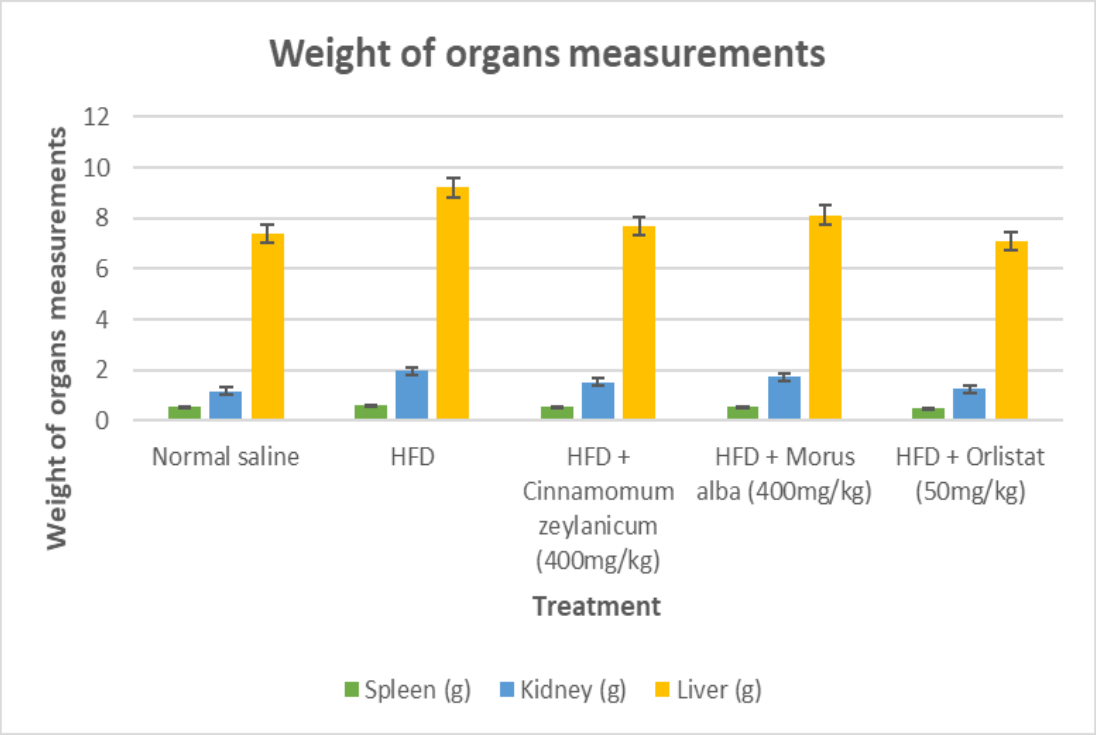

Fig. 2: Graphical data of weight of organs measurements.

Estimation of total cholesterol (TC)

After 15 days, HFD + *Cinnamomum zeylanicum* (400mg/kg) and HFD + *Morus alba* (400mg/kg) treated animals demonstrated the total cholesterol as 223.14±1.11mg/dl, and 137.46±1.36mg/dl, respectively.

Table 3: Estimation of Total Cholesterol.

Treatment	TC (mg/dl)				
	Day 1	Day 3	Day 5	Day 10	Day 15
Normal saline	165.46±0.34	163.45±1.62	166.35±1.26	162.12±1.23	163.48±1.23
HFD	241.12±1.44	243.45±1.12	244.22±1.56	247.20±1.47	249.46±1.68
HFD + *Cinnamomum zeylanicum* (400mg/kg)	216.21±1.46	212.45±1.54	236.11±1.35	234.14±1.24	223.14±1.11
HFD + *Morus alba* (400mg/kg)	227.24±1.12	230.24±1.14	131.43±1.43	134.31±1.45	137.46±1.36
HFD + Orlistat (50mg/kg)	173.32±1.11	178.22±1.34	179.30±1.45	182.10±1.17	183.11±1.56

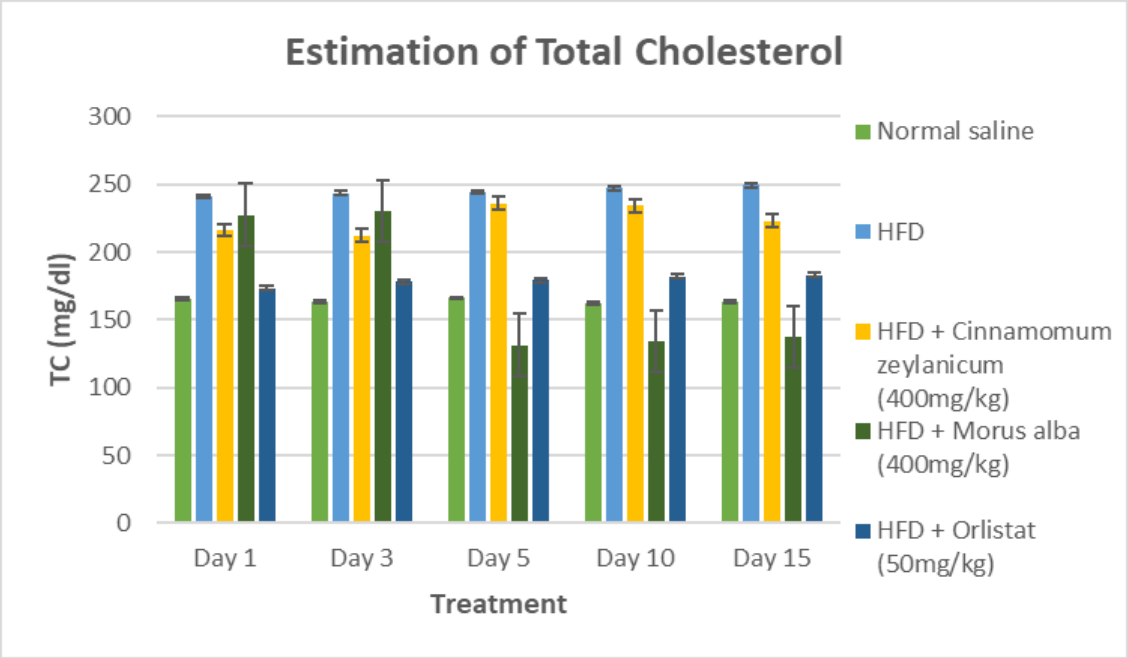

Fig. 3. Graphical data of Estimation of Total Cholesterol.

Food consumption pattern estimation

It was estimated at day 1, 3, 5, 10 & 15. HFD + *Cinnamomum zeylanicum* (400mg/kg) and HFD + *Morus alba* (400mg/kg) showed the effect in the ascending order, i.e., 96.34±0.12mg/dl and 23.12±0.12mg/dl, respectively. Both, the herbs exhibited a moderate increase in food consumption when compared with control.

Table 4: Food consumption pattern estimation.

Treatment	Food consumption pattern (mg/dl) Day 1	Day 3	Day 5	Day 10	Day 15
Normal saline	83.34±0.35	85.34±0.35	86.34±0.34	84.23±0.35	87.43±0.43
HFD	84.45±0.34	104.54±0.45	112.35±0.67	118.34±0.43	127.22±0.17
HFD + *Cinnamomum zeylanicum* (400mg/kg)	86.40±0.12	89.34±0.34	91.23±0.22	93.435±0.44	96.34±0.12
HFD + *Morus alba* (400mg/kg)	85.24±0.41	87.34±0.11	89.22±0.26	91.13±0.41	23.12±0.12
HFD + Orlistat (50mg/kg)	88.14±0.27	94.42±0.22	99.33±0.11	108.29±0.19	109.33±0.20

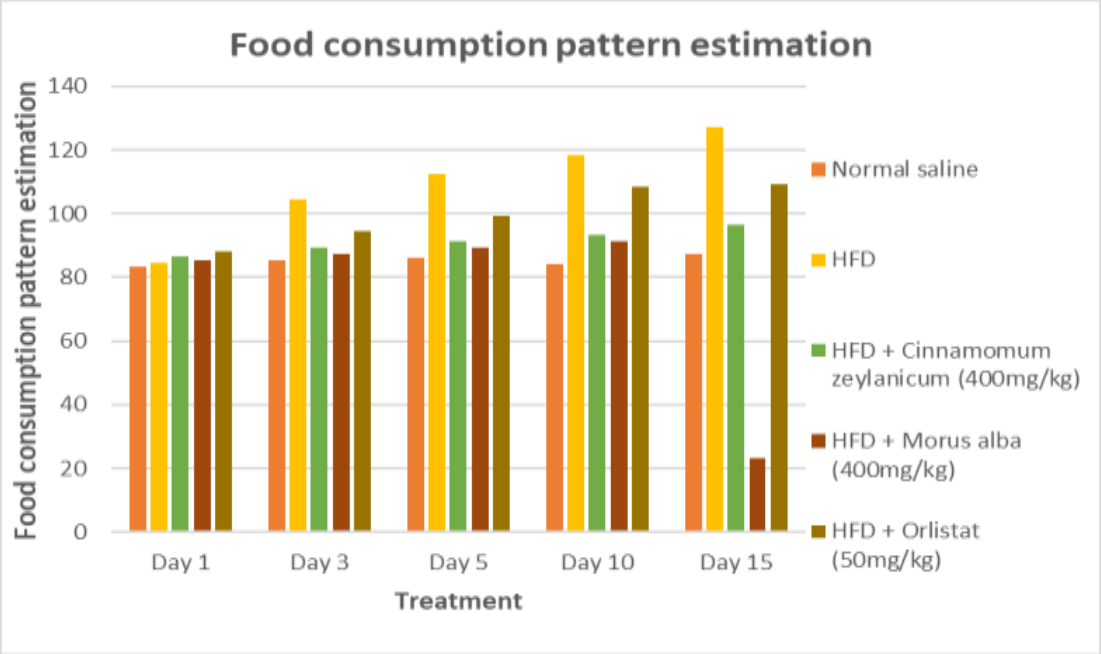

Fig. 4. Graphical data of Food consumption pattern estimation.

Estimation of blood sugar level

The blood sugar level was estimated at the day 1, day 3, day 5, day 10 and day 15. The HFD + *Cinnamomum zeylanicum* (400mg/kg) treated animals showed the blood glucose level as 90.43±0.42mg/dl, 92.20±0.23mg/dl and 94.20±0.34mg/dl, at the day 5, day 10 and day 15, respectively. However, HFD + *Morus alba* (400mg/kg) treated rats showed the blood glucose level as 91.45±0.64mg/dl, 94.46±0.25mg/dl and 96.20±0.12mg/dl, respectively.

These days, obesity lowers the standard of living. Lately, there has been a search for foods and nutrients with anti-obesity properties that come from natural sources. Research into Dendropanax morbifera's anti-obesity, cholesterol-lowering properties, and mode of action is, however, lacking.

Table 5: Estimation of blood sugar level.

Treatment	**Blood sugar level (mg/dl)**				
	Day 1	**Day 3**	**Day 5**	**Day 10**	**Day 15**
Normal saline	84.34±0.11	86.23±0.22	87.33±0.20	86.46±0.34	85.34±0.64
HFD	94.10±0.14	98.34±0.34	104.33±0.13	108.20±0.43	114.34±0.31
HFD + *Cinnamomum zeylanicum* (400mg/kg)	86.30±0.33	88.32±0.50	90.43±0.42	92.20±0.23	94.20±0.34
HFD + *Morus alba* (400mg/kg)	86.45±0.22	89.45±0.20	91.45±0.64	94.46±0.25	96.20±0.12
HFD + Orlistat (50mg/kg)	84.32±0.33	82.33±0.53	83.36±0.46	84.24±0.53	82.29±0.34

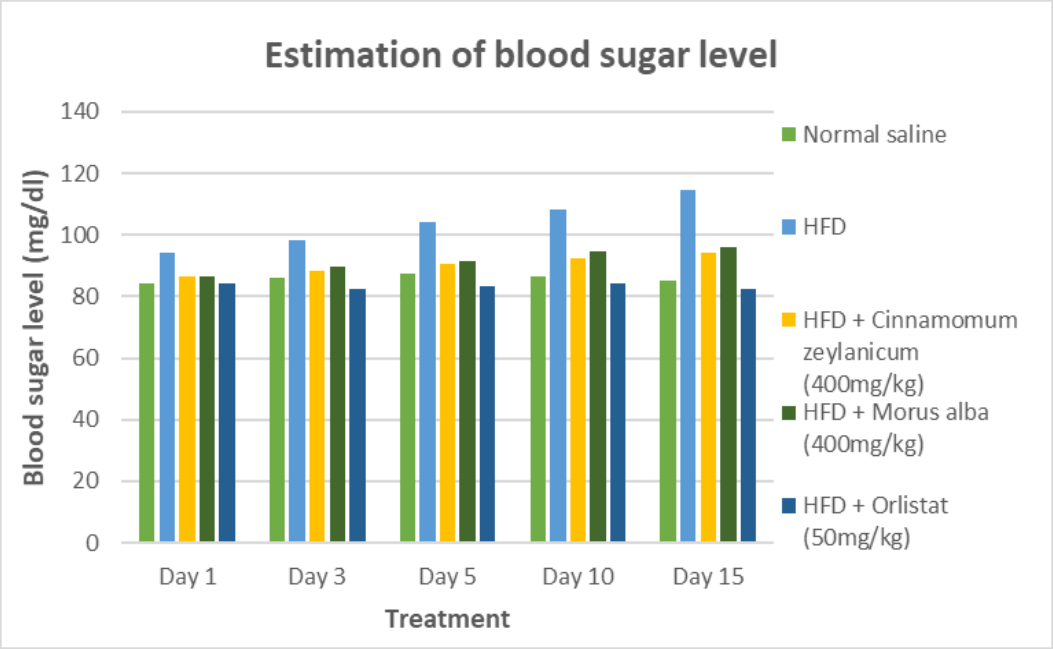

Fig. 5. Graphical data of Estimation of blood sugar level.

We have demonstrated that 3T3-L1 cells treated with DLW did not display cytotoxicity throughout a 24-hour period at DLW doses of 0–500 μg/mL. Furthermore, fat formation was markedly suppressed and the quantity of lipid droplets in 3T3-L1 cells treated with DLW for 3 to 9 days dropped. Furthermore, because DLW therapy inhibits adipogenesis during differentiation. As a result, Figure 1's toxicity was verified before D. morbifera's in vivo effectiveness was verified in mice (Lordan et al. 2011).

In biological tissue membranes, the lipid peroxidation reaction leads to the oxidative breakdown of polyunsaturated fatty acids by free radicals. MDA is utilized as an indicator, and a rise in it denotes an increase in oxidative stress. This consequently causes a decline in the membrane's ability to function, fluidity to diminish, and disturbance of homeostasis. It has been claimed that this is especially associated to senescent cells. This lowers the antioxidant defense capacity in vivo and increases the oxidative stress of cells in tissues. In this experiment, showed a 1.73, 2.03, 1.81, and 2.1-fold increase in SOD activity in comparison to the HF group. In addition, vascular endothelial cells emitted nitric oxide (NO). As a result, after receiving treatment for HF+DLW, mice's urine NO concentrations were considerably lower in the treated group ($p < 0.05$) than in the HF group (73.75 and 65.88%, respectively). TC content is a significant biomarker of lipid metabolism in the body (Childs et al. 2015). Therefore, DLW extract's impact on serum lipids may play a significant role in preventing obesity.

Phosphorylation activates AMPK, which is important for preserving energy balance. Additionally, AMPK inhibits lipid production and increases lipid oxidation. The expression of SREBP-1c is impacted and fat production is inhibited when phosphorylated AMPK activity rises. Consequently, the WAT of C57BL/6 obese mice validated the level of AMPK protein expression linked to the adipogenic process. A transcription factor that functions downstream of AMPK, SREBP-1c, is crucial for the synthesis of triglycerides in liver and adipose tissues (Jeon, 2016). In mitochondria, CPT1 is essential for the β-oxidation of lipids. We verified an increase in CPT1α/β in the HF+DLW500 and HF+GAR400 groups, respectively. Thus, when AMPK activation promotes phosphorylation of ACC (p-ACC) and ACC enzyme activity is decreased, fatty acid oxidation results from a reduction in malonyl-CoA (Foretz et al. 2018). Furthermore, as fatty acid oxidation increases, so does the quantity of ATP produced, which helps to reduce body fat (Li et al. 2011; Jones & Infante, 2015). To validate its anti-obesity potential, Cinnamomum zeylanicum and Morus alba (400 mg/kg) shown a significant modification in all the parameters. When compared to the control, it reduced the rats' body weights. Measurements of the weights of the kidneys, liver, and spleen revealed a decrease in body organ weight.

It is also associated with controlling total cholesterol and organ weights. It is evident to manage body weight as well as the gross weight of various organs when food consumption is stopped. The results showed a considerable reduction in blood sugar, total cholesterol, body weight, and organ weight, as well as hypolipidemic potential and anti-obesity.

CONCLUSION

When estimated lipid profiles in terms of total cholesterol, *Cinnamomum zeylanicum* and *Morus alba* demonstrated in lowering the biosynthesis of cholesterol thus maintained the level of total cholesterol. In the same order, triglyceride was also found decreased that may be because of inhibiting the synthesis of fatty acids and lipids. In an earlier investigation, Specifically, the HF+DLW500 group's ALT, AST activity, and MDA concentration dropped to the same levels as the control group. Additionally, NO generation was markedly suppressed in a number of HF+DLW groups. Therefore, use of *Cinnamomum zeylanicum* and *Morus alba* revealed that it might be significant in humans also to confer the anti-obesity and anti-hypercholesteremic effects after successful data in clinical trials. Therefore, based on all the variables and guidelines, it can be said that Cinnamomum zeylanicum and Morus alba are significant and potent anti-obesity drugs that should be administered to humans only after

clinical trials have shown the best safety and efficacy standards. Because of its widespread availability and mode of action, it would have a significant impact on the treatment of obesity, hypercholesterolemia, and high fat content.

This study suggests that *Cinnamomum zeylanicum* and *Morus alba* medication may be recommended as a well recognised treatment for obesity management and cholesterol reduction, thereby improving overall health and extending lifespan.

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Nil.

CONFLICT OF INTEREST

Authors declared for none conflict of interest.

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