



Efficacy of Trichilia Monadelpha Stem Bark Extracts on The Growth Performance of Growing Rabbits

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ABSTRACT

The utilization of medical plants is becoming more popular as a consequence of antibiotic resistance, the need to maintain a healthy environment, and the need to ensure the safety of food. Using a gas chromatography mass spectrometry (GC/MS) instrument, the secondary metabolites in trichilia monadelpha stem bark extract (TMSBE) were analyzed, and a Near Infra-red Spectrophotometer (NIR) kit was used to analyze the experimental diet's proximate components. There were 40 growing New Zealand white × Chinchilla crossbred rabbits of 6-7 weeks age (472.6 ± 6.2 g) were used for the experiment. Rabbits were randomly assigned to 4 groups; each treatment was further divided into 5 replicates consisting of 2 rabbits each. Group 1 (G1): Basal diet with no TMSBE, G2: Basal diet plus 3 mL TMSBE per rabbit/day, G3: Basal diet plus 6 mL TMSBE per rabbit/day and G4: Basal diet plus 9 mL TMSBE per rabbit/day. Experimental diet was formulated to meet the nutrient requirement standards for growing rabbits according to Nutritional Research Council standards (1977). GC/MS analysis of TMSBE revealed that it is abundant in copaene (25.40 %) followed by azulene (18.29 %), δ -cadinene (15.39 %), α -cubebene (10.61 %) and α -long pinene (10.11 %) respectively other compounds were less than 5 %. Average weekly weight gain (AWWG), average weekly feed intake (AWFI) and feed conversion ratio (FCR) were significantly ($P < 0.05$) different among the treatments. AWWG and AWFI follow similar trend and were highest in G3 and G4, intermediate in G2, lowest in G1 ($P < 0.05$). Better FCR were recorded among rabbits in G4 compared to the other groups ($P < 0.05$). Then, it can be concluded that feeding TMSBE at 9 ml/day improved feed intake and enhance performance in rabbits.

Keywords: Performance, Phytochemicals, Rabbits, Spectrophotometer, Trichilia Monadelpha

INTRODUCTION

Livestock farmers are transitioning to a production technique that uses few or no synthetic antibiotics at all in a new era. This is due to the increasing rate of antimicrobial resistance and more frequent notation of multidrug resistance strains (Amit, 2013). Environmental pollution, presence of toxic residue in the carcass leading to death of animal has also been identified as one of the disadvantages on indiscriminate use of antibiotics (J. O. Alagbe, 2022; Garg, 2011). The use of phytogetic (plant extracts) have been reported as the possible solution to promote food safety and boost livestock production especially after the European Union in 2006 placed a ban on the use of antibiotics in animal nutrition. Plant extracts contain phytochemicals which exhibit different biological properties in animal.

Phytochemicals are present in herbs, spices, and their extracts and serve a variety of purposes for plants, including pigmentation, growth, reproduction, disease resistance, and so on (Ankri & Mirelman, 1999; Sivropoulou et al., 1996). *In vitro* and *in vivo* studies have demonstrated the antibacterial, anti-cancer, anti-inflammatory, antimicrobial, anthelmintic, cytotoxic, immune-modulatory, hypolipidemic, hepato-protective and anti-oxidant benefits of these compounds (Shittu et al., 2022; Teissedre & Waterhouse, 2000). Examples of phytochemicals (secondary metabolites) are: tannins, flavonoids, terpenoids, saponins, alkaloids, phenols and they have been investigated as an effective nutritional strategy to improve feed intake and general performance of animals (Agubosi, Wika, et al., 2022; Bas, 2000). The chemicals concentrations of phytochemicals in plants varies from one plant species to another, other factors such as; storage conditions, location, climate as well as anti-nutrient could also play a key role (Agubosi, Alexander, et al., 2022; Liu et al., 2014). Among the potential plants with ethno-pharmacological or therapeutic properties is *Trichilia monadelpha*.

In the family of Maliaceae, *trichilia monadelpha* is one of the most important medicinal plant abundant in various phytochemicals of ethnopharmacological usefulness in livestock production. It is an evergreen semi-deciduous plant found mostly near river bank and widely distributed globally all over tropical and subtropical regions (Xie et al., 1994). The plant consist of over 90 species and it can grow to about 12 – 20 meters high with 0.4 m in girth with many biomedical properties (Pupo et al., 2002). *Trichilia monadelpha* roots, leaves and stem bark are traditionally used for the treatment of rheumatism, malaria, gastrointestinal tract, cough, arthritis, inflammation, and asthma and skin diseases. Gas chromatography mass spectrometry (GC/MS) of ethanolic extract of *Trichilia monadelpha* stem bark revealed the presence of Azulene, α -cubebene, β -caryophyllene, α -longipinene, γ -murolene, β -bisabolene, α -bergamotene, α -farnesene and β -cedrene having antimicrobial, antioxidant, cytotoxic, antianaphylaxis, analgesic, antiplasmodial, anti-inflammatory and immunomodulatory properties (Agarwal et al., 2006).

Scientific studies have also proven that aqueous extract from *trichilia monadelpha* stem bark, leaves and root are capable of inhibiting the activities of *E. coli*, *Salmonella spp*, *Staphylococcus spp* and other pathogenic microorganism making it a natural alternatives to antibiotics (Ben et al., 2013). Efficacy of *trichilia monadelpha* stem bark extract (TMSBE) have been tested on adult albino male rats, the outcome of the experiment revealed that the test material is capable of scavenging free radicals thus preventing disease and mortality in animals. However, there is scanty information on the impact of TMSBE on the general performance of growing rabbits.

The present study aimed to examine the impact of feeding different levels of *trichilia monadelpha* stem bark extract on the growth performance of rabbits as natural alternatives to synthetic antibiotics and to give a clue on its safety level.

RESEARCH METHODOLOGY

The research was conducted at Sumitra Research Institute, Gujarat, India. The test material used *Trichilia monadelpha* stem bark was collected at the Teaching and Research Farm, Sumitra Research Institute, India. The leaves were authenticated by a certified Taxonomist, he is Dr. Amit Shaka at the Department of Taxonomy, Sumitra Research Institute which had a saving number (MS/092/2021) which means the sample of the plant was deposited.

Extraction of *Trichilia Monadelpha* Stem Bark

In this section, we extracted the *trichilia monadelpha* stem bark by doing several ways, such as (1) in removing dust and other contaminants, the collected *Trichilia monadelpha* stem bark was first washed with running tap water and then with distilled water; (2) then, it was cut into smaller pieces with a kitchen knife to make it easier for the solvent to penetrate; (3) finally, it was allowed to air dry in a flat, clean metallic tray for 16 days before being ground into powder using a lab electric blender; (4) The Erlenmeyer flask was used to filter 100 g of *Trichilia monadelpha* stem bark powder after it had been dissolved in 500 ml of 95% ethyl alcohol for 24 hours while being stirred continuously; (5) In order to remove the solvent and recover the extract (TMSBE), the filtered liquid was placed in a water bath set at 4°C. It was then kept in a 4°C refrigerator until usage.

Animal Feeding, Health, Shelter, and Experimental Design

For the experiment, 40 growing New Zealand white x Chinchilla crossbred rabbits weighing 472.6 ± 6.2 g were chosen. To minimize stress, animals were obtained from a well-known farm in Gujarat and transported to Sumitra Research Institute in the early morning. After normalizing the weight and giving each rabbit an Ivomec® vaccine to remove potential parasites (endo- and ecto- parasites), the rabbits underwent a 2-week acclimation phase. The wire mesh-covered wooden hutches used to shelter the test animals were 150 cm from the ground. Four groups

of rabbits were randomly chosen, and each treatment was then divided into five replicates, each with two rabbits. An experimental diet was created to fulfill the nutritional parameters set by the Nutritional Research Council for growing rabbits (National Research Council, 1977). Since at start of the experiment, the animals were weighed to determine their initial body weight (IBW), which is represented in grams. The rabbits had the experimental diets twice a day at 7:00 am and 2:00 pm. The experimental design is a Completely Randomized Design, and biosecurity procedures were accorded the highest priority throughout the experiment's approximately 12-week period.

Data collection used Experimental Set-up

Daily feed leftovers and/or wastage were measured before supplying fresh feed, and daily feed consumption was monitored. Furthermore, fresh, clean water was constantly available during the experiment. Weekly body weight growth was recorded, in addition to average daily feed intake (calculated by the total feed intake by the variety of experimental days). Feed wastage and leftovers were subtracted from the total daily feed supplied to determine the daily feed intake. The ratio of feed intake to weight gain, or feed conversion ratio, was calculated while the mortality rate was tracked.

The experimental set-up was arranged such following below:

1. Group 1: Basal diet with no *Trichilia monadelpha* stem bark extract (TMSBE);
2. Group 2: Basal diet plus 3 mL TMSBE per rabbit/day;
3. Group 3: Basal diet plus 6 mL TMSBE per rabbit/day;
4. Group 4: Basal diet plus 9 mL TMSBE per rabbit/day.

Laboratory Analysis of *Trichilia Monadelpha* Stem Bark Extract Using Gas Chromatography Mass Spectrometry (GC-MS)

Using a SCION new generation Gas Chromatography Mass Spectrometry (GC-MS) system (GC-MS 436 Model) with an upper mass limit of 1200 m/z, programmable temperature vaporization injectors used in conjunction with "back flushing" to divert higher boiling point sample away from the column, high precision electronic pressure control, user created spectral libraries, and multiple spectral databases, *Trichilia monadelpha* stem bark extract (TMSBE) was subjected after turning on the power, 3 ml of TMSBE was injected into the machine's intake. Diffusion is prevented by the machine's glass gaps, and the installed TekLinK™ software was utilized to validate the results.

Using a Phoenix 5000 NIR feed analyzer with the following specifications: wave length (1100-2500 nm), dimensions (14.0 in x 15.0 in x 20.75 in) (LxWxH), and temperature, a chemical analysis of an experimental diet was conducted (35-105°F). Once the machine's switch was turned on, a dried sample of feed (200 g)

was put in the sample cap. Over less than 30 seconds, the machine automatically scans the whole feed surface and produces the results.

Statistical Analysis

All processes were compared using Turkey's test till the data were submitted to an ANOVA using SPSS (22.0), and effects were deemed significant $P < 0.05$.

Here the statistical model used:

$$Y_i = \mu + T_{ij} + e_{ij}$$

Description: Y_i = Effect on experimental observations,

μ = General mean of the population,

T_i = The effect of the dietary treatment,

e_{ij} = Error in the experiment.

Table 1.

Gross Composition of Experimental Diet Feed in Growing The Rabbits

Ingredient	Quantity (Kg)
Yellow Corn	24.00
Brewers dry grain	35.00
Palm kernel cake	14.00
Groundnut cake	6.60
Soy bean meal	15.00
Bone meal	2.00
Oyster shell	1.30
Salt	0.35
**Premix (Vitamins and minerals)	0.25
Toxin binder	0.20
Methionine	0.10
Lysine	0.20
Total	100.0
Determined analysis	
Dry matter (%)	91.22
Ash (%)	7.08
Crude protein (%)	16.10
Crude fiber (%)	15.40
Ether extract (%)	1.93
Digestible energy (Kcal/g)	280.18

** Each 1 kg contains: 7,500 IU vitamin A, 2621 IU vitamin D₃, 15.6 IU vitamin E, 2.10 mg vitamin K, 8.60 mg calcium pantothenate, 0.02 mg vitamin B₁₂, 0.55 mg folic acid, 300 mg choline chloride, 30.02 mg chlortetracycline, manganese 150.30 mg, 62.75 mg iron, 44.04mg zinc, 2.7 mg copper, 1.50 mg iodine, 0.34 mg cobalt, 0.11 mg selenium.

Table 2.

Analysis of Bioactive Compounds in *Trichilia Monadelpha* Stem Bark Extract Using GC-MS

Phytochemical Components	Reaction Time (min)	Area (%)	Molecular wgt (g/mol)
Copaene	2.81	25.40	204.3
α -santalene	4.52	0.02	203.8
Azulene	2.19	18.29	128.1
β -chamigrene	1.38	0.01	220.7
δ -cadinene	5.02	15.39	206.3
δ -selinene	9.27	0.44	205.2
β -elemene	13.10	0.02	194.3
α -cubebene	11.22	10.61	200.8
β -cayrophyllene	15.09	1.40	204.3
β -patcholene	10.30	0.49	204.4
α -longipinene	9.44	10.11	211.7
γ -murollene	10.29	2.50	154.3
α -pinene	15.25	1.67	188.5
γ -terpinene	19.01	0.77	180.6
γ -eudesmol	20.32	1.15	188.3
β -bisabolene	25.88	0.75	188.5
α -farnesene	24.30	1.04	171.4
β -lonupinene	27.01	0.71	200.9
α -bergamotene	27.02	0.22	196.3
Eicosane	29.14	1.93	192.7
β -cedrene	29.59	0.16	152.0
Aromadendrene	30.82	0.70	164.7
3-methoxy-p-cymene	30.52	1.23	154.2
α -Elemene	33.60	0.88	222.2
Sum total	-	95.89	-

Table 3.The Impact of *Trichilia Monadelpha* Stem Bark Extract on The Growing Rabbits

Variables	G1	G2	G3	G4	SEM
No of animals	10	10	10	10	-
Breed	NWZ ×CHIN	NWZ ×CHIN	NWZ ×CHIN	NWZ ×CHIN	-
IBW (g)	477.8	478.8	473.2	472.6	0.02
AWLW (g)					
5 – 6	531.6	522.9	530.4	529.4	10.91
6 – 7	728.1	743.8	750.7	755.2	12.33
7 – 8	891.2	900.8	923.6	940.4	13.04
8 – 9	1066.2 ^b	1137.1 ^a	1220.1 ^a	1228.7 ^a	18.70
9 -10	1102.2 ^c	1334.1 ^b	1502.5 ^a	1511.4 ^a	19.75
10 -11	1206.1 ^b	1457.2 ^a	1669.3 ^a	1802.8 ^a	15.03
11-12	1390.6 ^c	1511.0 ^b	1788.8 ^b	1924.7 ^a	12.48
AWWG (g)					
5 – 6	128.5 ^c	120.4 ^b	169.0 ^a	170.6 ^a	6.83
6 – 7	173.1 ^c	195.8 ^b	201.1 ^a	218.7 ^a	7.54
7 – 8	196.0 ^b	211.8 ^a	256.7 ^a	266.2 ^a	8.06
8 – 9	196.5 ^c	298.3 ^b	312.4 ^a	318.4 ^a	5.15
9 -10	163.1 ^c	306.2 ^b	408.7 ^a	433.1 ^a	3.86
10 -11	136.0 ^c	356.3 ^b	471.8 ^a	480.2 ^a	4.97
11-12	114.5 ^c	381.1 ^b	588.5 ^a	597.1 ^a	5.08
AWFI (g/w)					
5 – 6	614.8	618.7	620.6	622.4	9.12
6 – 7	770.0	764.2	766.8	767.2	6.17
7 – 8	805.4	811.7	810.4	809.1	8.32
8 – 9	847.1	852.9	860.1	859.6	6.51
9 -10	875.3	880.4	879.2	880.1	5.16
10 -11	896.5 ^b	898.7 ^b	900.1 ^a	997.4 ^a	7.47
11-12	712.1 ^c	890.5 ^b	918.7 ^a	923.5 ^a	6.02
FCR					
5 – 6	5.10 ^a	4.93 ^b	3.67 ^c	3.64 ^c	0.96
6 – 7	4.45 ^a	3.90 ^b	3.81 ^b	3.51 ^c	0.60
7 – 8	4.11 ^a	3.83 ^b	3.19 ^c	3.04 ^d	0.51
8 – 9	4.31 ^a	3.01 ^b	2.99 ^c	2.91 ^c	0.42
9 -10	5.36 ^a	2.90 ^b	2.88 ^b	2.71 ^c	0.21
10 -11	5.91 ^a	2.87 ^b	2.61 ^c	2.60 ^c	0.10
11-12	4.93 ^a	3.00 ^b	2.99 ^c	2.90 ^c	0.15
Overall AWWG (g)	912.8 ^c	1032.2 ^b	1315.6 ^a	1452.1 ^a	25.87
Overall AWFI (g/w)	865.20 ^c	871.72 ^b	900.34 ^a	901.16 ^a	21.04

RESULT AND DISCUSSION

The Analysis of Bioactive Compounds in *Trichilia Monadelpha* Stem Bark (TMSBE) Extract Using GC-MS

Phytochemicals also known as bioactive chemicals, are organic compounds of a plant origin that are used by plants for growth and pathogen defense. They also have a significant impact on the sensory qualities (color, taste, and fragrance) of plants (Shittu & Alagbe, 2020). Azulene, copaene, and δ -cadinene, which contain 25.40%, 18.29%, and 15.39 percent of the total 24 compounds identified in the GC-MS analysis of *Trichilia monadelpha* stem bark extract, respectively, are phenolic compounds with one or more aromatic rings and one or more hydroxyl groups. These compounds exhibit various biological properties in livestock production and health. The second most abundant compounds in TMSBE are α -cubebene (10.61%) and α -longipinene (10.11%), which belong to a group of flavonoids with antimicrobial, antiviral, and antioxidant properties. Phenolic compounds have antioxidant and anti-inflammatory properties and the ability to inhibit the growth of pathogenic bacteria (J. Alagbe, 2021). Naturally, flavonoids often exist as compounds that are glycosylated or esterified, while they can also change into aglycones during the extraction process (Adewale et al., 2021) (Agubosi, Soliu, et al., 2022). Other compounds [α -santalene (0.02%), β -chamigrene (0.01 %), δ -selinene (0.44 %), β -elemene (0.02 %), β -cayrophyllene (1.40 %), β -patcholene (0.49 %), γ -murollene (2.50 %), α -pinene (1.67 %), γ -terpinene (0.77 %), γ -eudesmol (1.15 %), β -bisabolene (0.75 %), α -farnesene (1.04 %), β -lonupinene (0.71 %), α -bergamotene (0.22 %), eicosane (1.93 %), β -cedrene (0.16 %), aromadendrene (0.70 %), 3-methoxy-p-cymene (1.23 %) and α -elemene (0.88 %)] obtained during the GC-MS analysis of TMSBE were less than 5 %. However, it has a variety of therapeutic characteristics, including the ability to scavenge reactive or poisonous substances, act as an enzymatic component, suppress pathogenic intestinal flora, and serve as a substrate for biochemical processes (J. Alagbe, 2021). The finding of this research on the analysis of bioactive components in *trichilia connaroides* leaf extract correlate with that of Ravendra (2011) but differ from Purnima (2006). The different results contribute to the type of extraction used, the age of the plant, variations in the location and species (Akintayo and Alagbe, 2021; Musa et al., 2021).

The Impact of *Trichilia Monadelpha* Stem Bark Extract on The Growing Rabbits

Table 3 provides data on the impact of *trichilia monadelpha* stem bark extract on growing rabbits. The average weekly weight gain in grams (g) per rabbit ranges from 128.5-170.6 g on the fifth and sixth week to 173.1-218.7 g, 196.0-266.2 g, 196.5-318.4 g, 163.1-433.1 g, 136.0-480.2 g, and 114.5-597.1 g on the sixth and seventh week to the eighth and ninth weeks, the tenth and eleventh weeks, and the twelfth and twelveth weeks, respectively. The G3 and G4 groups had the greatest

overall AWWG, accompanied by G2 and G1, respectively ($P < 0.05$). An indication of enhanced enzymatic activity in the intestinal system and a resultant increase in the absorption of nutrients from the food consumed to the rabbits is the larger weekly weight gain in G3 and G4 compared to other groups. The effects of phytochemicals on enhancing animal productivity include activating internal secretions, boosting animal hunger, and inhibiting the actions of dangerous microorganisms (Ahmed, 2000). The results indicate that feeding growing rabbits 6–9 ml of TMSBE daily has an advantage over the other treatments in terms of improving animal productivity ($P < 0.05$). It might also indicate that the animals fed TMSBE kept their guts in good condition and will have the best possible health and productivity. The average weekly feed intake (AWFI) was found to differ significantly ($P < 0.05$) across the groups. The range of the average weekly feed intake was 865.20–901.16 g. Additional food was eaten by the rabbits in G3 and G4 compared to the other groups, which resulted in higher FCRs. This experiment's result is consistent with Dalle and Attia observed when they fed growing rabbits phytogenics.

CONCLUSION

Because they include phytochemicals with a variety of therapeutic qualities, plant extracts exhibit a larger range of animal feeding activities than synthesized drugs. They are safer for the environment and less harmful. According to the findings of the research, feeding growing rabbits up to 9 ml of *trichilia monadelpha* stem bark extract per day not have a negative impact on their growth.

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