



Nutritional Composition of Ten Ethnobotanicals Used for the Treatment of Anaemia in Southwest Nigeria

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ABSTRACT

Aim: Ethnobotanical investigation revealed that *Parquetina nigrescens*, *Sorghum bicolor*, *Terminalia catappa*, *Trema orientalis*, *Mangifera indica*, *Waltheria indica*, *Theobroma cacao*, *Harungana madagascariensis*, *Tetracera alnifolia* and *Detarium microcarpum* are used traditionally for the treatment of anaemia in southwestern Nigeria. This study screened the plants for their proximate constituents and phytochemical compounds to provide scientific details for their therapeutic use for the treatment of anaemia.

Study design: Proximate and phytochemical analyses of ten ethnobotanicals.

Place and Duration of Study: Departments of Botany, Pharmacognosy and Animal Nutrition, University of Ibadan, between January and September, 2010.

Methodology: Proximate and phytochemical analyses of plant parts of ten ethnobotanicals were carried out using standard laboratory methods. Data were analysed using Statistical Analysis System (SAS). Differences between means were assessed for significance at $p < 0.05$ by Duncan's Multiple range test (DMRT).

Results: The habits of the tested plants were 60% trees, 30% shrubs and 10% herbs. The use-value of plant parts were 60% barks and 40% leaves. The highest value (19.95%) of crude protein was recorded for *P. nigrescens*. *S. bicolor* showed significantly ($P < 0.05$) high content of crude fibre (30.00%) and highest dry matter was obtained from *T. cacao* and *T. catappa*. Anthraquinones were present in *Harungana madagascariensis*, *Theobroma cacao*, *Mangifera indica* and *Waltheria indica*, 70% of the test plants contained tannins, and cardiac glycosides were present in all plant samples. This study, thus confirms the nutritional potential of the test plants in addition to their active phytochemical constituents. Their nutrients might complement the active phytochemicals in therapeutic activities.

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Conclusion: It was concluded that there is need for the isolation and identification of the active compounds responsible for their antianaemic activities. Furthermore *P. nigrescens*, *M. indica* and *T. cacao* could be used as food supplements in weaning food because of their significant crude protein and fibre constituents in addition to their therapeutic potential.

Keywords: Ethnobotany; haematinics; medicinal plants; proximate analysis; phytochemical screening; Nigeria.

1. INTRODUCTION

Anaemia is a common blood disorder that affects people of all ages although the people at greater risk are the elderly, young women of child bearing age and infants. This condition is not a disease but could develop as a result of various diseases (WHO and UNICEF, 2001). There are over 400 types of anaemia, many of which are rare but in all cases there is lower than normal number of circulating red blood cells (Ogbe et al., 2010). The most common form of anaemia is caused by a deficiency of iron which is an essential constituent of haemoglobin. The main cause of iron deficiency anaemia is iron loss due to heavy or persistent bleeding and menstruation is the most common cause in women of child bearing age. Other causes include blood loss from the digestive tract due to disorders such as erosive gastritis, peptic ulcer, stomach ulcer, inflammatory bowel disease, hemorrhoids, and blood tumours (Illustrated Medical Dictionary, 2002). Iron deficiency is the most common cause of nutritional anaemia which affects over 600 million people throughout the world particularly in developing countries (Oladiji et al., 2003).

In some countries, infectious diseases such as hookworms, ascaris, and schistosomiasis, also acute and chronic infections including malaria, cancer, tuberculosis and HIV/ AIDS are important factors contributing to the high prevalence of anaemia in many populations. For example, *Plasmodium falciparum* malaria related anaemia contributes significantly to maternal and child mortality and thus prevention and treatment of anaemia in pregnant women and young children are of major importance (Staubli, 2001). Iron deficiency anaemia was considered to be among the important contributing factors to the global burden of diseases (Gadaga et al., 2009).

Sickle cell anaemia is one of the diseases ravaging most world population cutting across nations and ethnic divide. According to reports, Africa is believed to be the origin of sickle cell anaemia and those afflicted with the disease are estimated at 200,000 per year. The recurrent and painful symptoms experienced during crises by sickle cell patients are known by various names in different parts of the world with complaints of shortness of breath, heart palpitations, abdominal pains, aches and pains in the muscle (Dapa and Gill, 2002).

Although there are various drugs used for the treatment of anaemia, they are not affordable to many poor people in the developing countries such as Nigeria. In addition the rural populations in various parts of the world do not have adequate access to high quality drugs for the treatment of anaemia, so depend heavily on plants and herbal products for the treatment of diseases and anaemia. Anaemia is claimed to have been successfully treated with plant materials by traditional medicine practitioners and many authors (Burkill, 1985;

Okpuzor et al., 2008). The extract of *Pterocarpus santalinoides* and *Aloe vera* was reported to increase the gelling time of sickle cell blood and inhibits sickling *in-vitro*. This indicates that such plants may be useful in the management of sickle cell disorder (Okpuzor et al., 2008). The reversal of sickling by use of medicinal plants has been reported: *Terminalia catappa* could be an effective antisickling agent inhibiting osmotically induced haemolysis of human erythrocytes (Mgbemene and Ohiri, 1999). Phytomaterials such as *Annona senegalensis*, *Cymbopogon densiflorus*, *Bridelia ferruginea*, *Ceiba pentandra*, *Morinda lucida* and *Alchornea cordifolia* have been reported to be useful in the treatment of anaemia (Mpiana et al., 2007). The role of crude aqueous extract of *Zanthoxylum macrophylla* roots as an antisickling agent was also highlighted and 2-hydroxybenzoic acid was isolated and identified as the antisickling agent obtained from the root of this plant (Elekwa et al., 2005). An investigation on the aqueous extracts of *Garcinia kola* confirmed that it could be useful in the management of anaemic diseases (Elekwa et al., 2003).

Parquetina nigrescens (Afzel.) Bullock, *Sorghum bicolor* (Linn.), *Terminalia catappa* (Linn.), *Trema orientalis* (Willd.), *Mangifera indica* (Linn.), *Waltheria indica* (Linn.), *Theobroma cacao* (Linn.), *Harungana madagascariensis* (Lam. ex Poir.), *Tetracera alnifolia* (Linn.) and *Detarium microcarpum* (Guill. and Perr.) are commonly used in the treatment of anaemia in southwestern Nigeria. This study screened the ten ethnobotanicals for their proximate and phytochemical constituents to provide scientific details accountable for their traditional use as anti-anaemia.

2. MATERIALS AND METHODS

2.1 Ethnobotanical Investigation

The informal ethnobotanical investigation was conducted in Yoruba language. The respondents were female herb-sellers in three local herbal markets in Ibadan. They were questioned on their traditional knowledge of the treatment of anaemia. Recipes were documented. The local name, parts of plant used, method of preparation and mode of administration were also recorded (Sofowora, 1993).

2.2 Collection and Identification of Plant Materials

Fresh and healthy plant materials were used for this study. The plant-parts of *Sorghum bicolor*, *Harungana madagascariensis*, *Detarium microcarpum*, *Tetracera alnifolia* were purchased from a local herbal market (Bode) in Ibadan, Nigeria while those of *Parquetina nigrescens*, *Theobroma cacao*, *Trema orientalis*, *Mangifera indica*, *Terminalia catappa* and *Waltheria indica* were collected from the nursery and botanical garden of University of Ibadan. The voucher specimens of test plants were identified and deposited in the University of Ibadan Herbarium (UIH).

2.3 Preparation of Plant Materials

The plant materials were washed thoroughly and air dried in the laboratory at room temperature for four weeks, each plant sample was milled to a coarse powder and stored in a glass container for further use.

2.4 Proximate Analysis of Plant Samples

The proximate analysis of the powdered plant samples for protein, fat, fibre, ash and dry matter was determined using the methods described in AOAC (1990) at the Department of Animal Nutrition, Faculty of Agricultural Science, University of Ibadan.

2.4.1 Determination of dry matter of plant samples

The plant sample was thoroughly mixed with water in a bottle. The water content was determined by weighing out 2 g of the sample into a silica dish which has been previously ignited and weighed, it was dried in the oven for 24 hrs at 100°C, and it was then allowed to cool for 10 minutes in a desiccator before weighing.

$$\% \text{ moisture (residual)} = \frac{\text{wt. of sample taken} - \text{wt. of sample after drying}}{\text{wt. of sample}} \times 100$$

Dry matter = 100 - % of moisture.

2.4.2 Determination of ash of plant samples

The residue from the moisture was charred over a flame and the furnace was ignited until the ash was grey, it was allowed to cool and then weighed.

2.4.3 Determination of ether extract (oil) of plant samples

A Soxhlet extractor was fixed with a reflux condenser and a small flask which has been previously dried in the oven and weighed; 1 g of sample was weighed and transferred to a fat free extraction thimble which was plugged lightly with cotton wool. Then the thimble was placed in the extractor and petroleum ether was added, once it siphoned more ether was added until the barrel of the extractor is half full. The ether boils gently and it was left to siphon ten times, then the flask was detached and the content was poured into the ether stock bottle. The condenser and the flask were replaced and the ether was distilled until the flask was dry. The flask which now contained all the oil was detached; the oil was cooled and then weighed.

$$\% \text{ ether extracts} = \frac{\text{wt. of oil}}{\text{wt. of sample}} \times 100$$

2.4.4 Determination of crude protein of plant samples

2 g of the sample was weighed into a Kjeldahl flask, 5 g of anhydrous sodium sulphate and 25 ml of concentrated H₂SO₄ were added to it. Thereafter it was placed in the fume cupboard and heated gently for 5 - 10 minutes after which frothing have nearly ceased and the solution gives a green colouration. It was allowed to cool and was diluted with water, the % N₂ in the sample was determined using the micro Kjeldahl apparatus.

2.4.5 Determination of crude fibre of plant samples

25 ml of 10 % sulphuric acid was measured with a pipette into a beaker and 175 ml of water was added while the residues from the ether extract was added and allowed to boil. When the liquid had boiled for exactly 30 minutes, it was poured into the funnel and filtered by

suction. The residue was washed with hot water until it was free from acid, then the residue was turned into a digesting flask and 200 ml of 1.25 % sodium hydroxide solution which was previously boiled was added. It was then filtered through a Whatman No.4 filter paper and the whole solution was poured into the filter and washed with boiling water and 1 % hydrochloric acid until it was free from acid. The residue was washed twice with 95 % alcohol and three times with petroleum ether using small quantities; the residue was allowed to drain and was transferred into a silica dish. It was thereafter dried in the oven to remove all organic matter and weighed after cooling.

2.5 Phytochemical Screening

The phytochemical screening of the samples was carried out using in the Department of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Nigeria. The steps taken for the phytochemical screening are in line with standard procedures (Harborne, 1984; Sofowora, 1993 and Evans 1996).

2.5.1 Alkaloids

500 mg of the powdered plant sample was weighed and extracted with 10 ml of 2 % hydrochloric acid (HCl). The HCl extract was then filtered with Whatman filter paper (No.1) so as to have a clear solution and also to prevent false results. The filtrate of about 2.5 ml was treated with few drops of Dragendoff's reagent. A precipitate indicated the presence of alkaloids.

2.5.2 Tannins

500 mg of the sample was mixed with 10 ml of distilled water and heated on water bath. The mixture was filtered and ferric chloride (FeCl_3) was added to the filtrate. Appearance of blue-black colouration showed the presence of tannins.

2.5.3 Saponins

200 mg of the sample was shaken with 5 ml of distilled water and then heated to boil. Persistent frothing showed the presence of saponins.

2.5.4 Cardiac glycosides

500 mg of the sample was dissolved in 2 ml of glacial acetic acid containing one drop of FeCl_3 . The solution was underplayed with 1 ml of concentrated sulphuric acid (H_2SO_4). A brown ring at the interphase between the acetic acid layer and H_2SO_4 layer indicated the presence of cardiac glycosides (Keller-Killiani's test).

2.5.5 Anthraquinones

500 mg of the sample was shaken with 10 ml of benzene. The solution was filtered and 5 ml of 10% ammonium hydroxide (NH_4OH) solution was added to the filtrate. A violet colour was observed in the lower phase. It indicated presence of anthraquinones.

2.6 Statistical Analysis

Analysis of variance and comparison of means were carried out on all data of proximate analysis of the plant samples using Statistical Analysis System (SAS). Differences between means were assessed for significance at $p < 0.05$ by Duncan's Multiple range test (DMRT).

3. RESULTS AND DISCUSSION

3.1 Ethnobotanical Investigation and Profile of Plants Used in This Study

The indigenous recipes are presented in Table 1. In the ethnobotanical investigation, the extraction of plant materials in alcohol and charring of plant materials were not recorded. Preparation of decoction from plant materials formed the most common method of preparation in remedies for anaemia. The preference in preparation method may depend on potency of the herbal remedy. Interestingly, given dosages or metric measures is part of the culture of respondents encountered and no incantation was recorded in this work. Furthermore the use of spices such as *Xylopiya aethiopica* and *Aframomum melegueta* as part of recipes was documented. The two spices may serve as immune booster and in the improvement of blood circulation.

Table 1. Herbal recipes for the treatment of anaemia in southwest Nigeria

S/N	Herbal combination and dosage	Method of preparation
1.	The barks of <i>Detarium microcarpum</i> and <i>Harungana madagascariensis</i> are cut into pieces, dried and ground into powder. One teaspoonful of the powder is taken with hot pap once daily.	Powder
2.	The leaves of <i>Sorghum bicolor</i> and the bark of <i>Theobroma cacao</i> are boiled with water and 250 ml of the preparation is taken twice daily.	Decoction
3.	The leaves of <i>Parquentina nigrescens</i> should be squeezed with water and 150 ml of the juice should be taken twice daily.	Infusion
4.	<i>Waltheria indica</i> leaves should be squeezed with water and little honey is added to the preparation. 150 ml of the preparation is taken once daily.	Infusion
5.	The bark of <i>Mangifera indica</i> and small quantity of <i>Aframomum melegueta</i> fruits are dried and ground into powder. One tablespoonful of the powder is taken once daily.	Powder
6.	Fallen leaves of <i>Terminalia catappa</i> and leaves of <i>Parquentina nigrescens</i> are boiled in water. 250 ml of the juice is taken once daily.	Decoction
7.	The bark of <i>Theobroma cacao</i> is boiled with water and mixed with hot pap as baby food.	Concoction
8.	<i>Xylopiya aethiopica</i> fruits, <i>Trema orientalis</i> bark, <i>Tetracera alnifolia</i> bark, <i>Trema orientalis</i> bark and <i>H.madagascariensis</i> bark are boiled with water. 150 ml of the preparation is taken twice daily.	Decoction
9.	<i>Sorghum bicolor</i> leaves are cooked with beans in water as food.	Concoction
10.	Leaves of <i>Terminalia catappa</i> and <i>Waltheria indica</i> are boiled with water and 250 ml is taken once daily.	Decoctions

The profile of plant samples used in this study is presented in Table 2. The habits of test plants showed that trees are mostly used which is an implication that further search for anti-anaemic plants should include higher medicinal plants. The use-value of plant parts are mostly barks (60%), the barks might be very rich in active phytochemical compounds and valuable nutrients (Table 2). The collection of plant barks for therapeutic purposes has implication on conservation of plant genetic resources, therefore the indigenous people should be enlightened on the need for conservation of over- exploited and endangered medicinal plants.

Table 2. Profile of plants used as heamatinics in yoruba ethnomedicine, Nigeria

Botanical name	Family	Local name (Yoruba)	Plant habit	Part used
<i>Mangifera indica</i>	Anacardiaceae	Mangoro	Tree	Bark
<i>Terminalia catappa</i>	Combretaceae	Afara	Tree	Leaves
<i>Tetracera alnifolia</i>	Dilleniaceae	Opon	Tree	Bark
<i>Waltheria indica</i>	Sterculiaceae	Ewe epo	Shrub	Leaves
<i>Theobroma cacao</i>	Sterculiaceae	Koko	Tree	Bark
<i>Trema orientalis</i>	Ulmaceae	Afere	Tree	Bark
<i>Parquetina nigrescens</i>	Periplocaceae	Ewe ogbo	Shrub	Leaves
<i>Sorghum bicolor</i>	Poaceae	Oka baba	Herb	Leaves
<i>Harungana madagascariensis</i>	Hypericaceae	Amuje	Shrub	Bark
<i>Detarium microcarpum</i>	Caesalpinaceae	Arira	Tree	Bark

3.2 Proximate Analysis of Plant Samples

Table 3 shows the proximate content of plant samples. The % yield of crude protein (19.95%) in *Parquetina nigrescens* was significantly high, there was no significant ($P < 0.05$) difference between the values of protein content of *Sorghum bicolor* and *Detarium microcarpum* and the least value (1.23%) was recorded for *Mangifera indica* and *Tetracera alnifolia*. The % yield of crude fibre of *Sorghum bicolor* (30.00 %) was significantly high and there was no significant difference between the values of crude fibre of *Parquetina nigrescens*, *Terminalia catappa*, *Detarium microcarpum* and *Trema orientalis*, the least values (11.00 %) was recorded for *Harungana madagascariensis* and *Waltheria indica*. *Parquetina nigrescens* was significantly high in ash content (9.00 %) when compared to other samples: there was no significant difference between the ash values of *Theobroma cacao*, *Terminalia catappa* and *Magnifera indica*. The least value of ash (4.00%) was recorded for *Harungana madagascariensis* and *Sorghum bicolor*. The proximate analysis showed varied values of nutrient composition in the samples. The results of this study conform to the findings of other authors with variation in the values of the proximate constituents due to differences in plant- parts used in the various studies: The relatively high crude fibre content in *W. indica* will help to maintain the movement of food through the gut and may be broken down by enzymes and bacteria in the gut to provide energy (Oladiji et al., 2005). The fruit pulp of *D. microcarpum* can serve as a source of foods for man and livestock because of its crude protein, crude fibre and carbohydrate content (Obun et al., 2010a). Mariod et al. (2009) analysed *D. microcarpum* fruit for its proximate compositions and recorded 29.1 - 30.9 % crude protein in its dried fruit pulp. Nzikou et al. (2010) studied the extraction and characteristics of seed kernel oil from *M. indica* and recorded that the seeds kernels have ash content of 3.2 %. Imaga et al. (2010) reported that *P.nigrescens* has appreciable antisickling activity; it has no toxic effect when administered at low

concentrations and protect the integrity of the erythrocyte membrane as evidenced in the hemolysis of the Hbss cells. Adebisi et al. (2005) recorded 21.00 % crude protein content in *S. bicolor* starch hydrolyzed with amylase from *rhizopus* sp. The administration of aqueous extract of *S. bicolor* stem bark in rats restored the anaemic condition in the iron deficient group and thus lends credence to its use in folklore medicine in the management of anaemia (Oladiji et al., 2007). Matos et al. (2009) evaluated the composition and nutritional properties of seeds and oil from *T. catappa*. The seeds of *T. catappa* contained 13 % moisture, 23.78 % crude protein, 4.27 % ash, and 4.94% crude fibre. Chung et al. (2003) studied the compositional characterization of *T. cacao* and recorded 93 g/kg ash content in *T. cacao* hull. There is dearth of information in literature on the proximate analysis of *T. alnifolia*, *T. orientalis* and *H. madagascariensis*.

Table 3. Results of the proximate analysis of powdered plant samples

Sample	Crude Protein (%)	Crude fibre (%)	Ether extract (%)	Ash (%)	Dry matter (%)
<i>Mangifera indica</i>	*1.23 ^f ± 1.00	24.00 ^{ba} ± 1.00	7.00 ^a ± 1.00	6.00 ^{dc} ± 1.00	91.50 ^a ± 10.00
<i>Terminalia catappa</i>	12.60 ^b ± 1.00	18.00 ^{bc} ± 10.00	12.00 ^a ± 10.00	6.00 ^{dc} ± 1.00	93.50 ^a ± 10.00
<i>Tetracera alnifolia</i>	1.23 ^f ± 1.00	25.00 ^{ba} ± 1.00	7.00 ^a ± 1.00	5.00 ^{de} ± 1.00	91.00 ^a ± 10.00
<i>Waltheria indica</i>	11.90 ^b ± 1.00	11.00 ^c ± 0.00	8.00 ^a ± 1.00	5.00 ^{de} ± 1.00	89.00 ^a ± 10.00
<i>Theobroma cacao</i>	3.85 ^{ed} ± 1.00	22.00 ^{ba} ± 10.00	9.00 ^a ± 1.00	6.00 ^{dc} ± 1.00	93.50 ^a ± 10.00
<i>Trema orientalis</i>	4.20 ^d ± 1.00	13.00 ^{bc} ± 1.00	12.00 ^a ± 10.00	8.00 ^{ba} ± 1.00	89.00 ^a ± 10.00
<i>Parquetina nigrescens</i>	19.95 ^a ± 1.00	14.00 ^{bc} ± 10.00	12.00 ^a ± 10.00	9.00 ^a ± 1.00	90.50 ^a ± 10.00
<i>Sorghum bicolor</i>	4.90 ^c ± 1.00	30.00 ^a ± 10.00	11.00 ^a ± 0.00	4.00 ^e ± 1.00	92.50 ^a ± 10.00
<i>Harungana madagascariensis</i>	2.28 ^{ef} ± 1.00	10.00 ^c ± 0.00	10.00 ^a ± 0.00	4.00 ^e ± 1.00	88.00 ^a ± 10.00
<i>Detarium microcarpum</i>	7.88 ^c ± 1.00	13.00 ^{bc} ± 1.00	9.00 ^a ± 1.00	7.00 ^{bc} ± 1.00	90.50 ^a ± 10.00

*Mean of 3 readings ± standard deviation.

*Different letters in the same column indicate significant differences ($p < 0.05$).

3.3 Phytochemical Screening of the Plant Samples

The phytochemical screening is presented in Table 4. Anthraquinones were present in *Harungana madagascariensis*, *Theobroma cacao*, *Magnifera indica* and *Waltheria indica*, but absent in *Parquetina nigrescens*, *Terminalia catappa*, and *Trema orientalis*. Alkaloids were present in *Parquetina nigrescens*, *Magnifera indica* but absent in *Harungana madagascariensis*, *Tetracera alnifolia* and *Waltheria indica*. Saponins were present in 90 % of the samples. Tannins were absent in *Sorghum bicolor* and *Tetracera alnifolia* but present in other samples. Cardiac glycosides were present in all samples. The results of the

phytochemical analysis agree with the findings of other authors: Yakubu et al. (2005) reported that the presence of saponins, phenolics and glycosides may be responsible for the acclaimed anti-anaemic potential of plants used in traditional medicine. Saponins especially are known to enhance natural resistance and recuperative powers of the body (Singh *et al.*, 1991). The occurrence of tannins shows that the plants may be useful in various ways for example tannins are useful in food and pharmaceutical (Babayi et al., 2004). Obun et al. (2010b) evaluated *D. microcarpum* pulp meal as feed ingredient in the diets of growing rabbits and reported that it contained low level of tannins (0.023 %), however it has impressive performance nutrient digestibility and economically viable alternative basal feed stuff to maize. Kumar et al. (2009) reported that the preliminary phytochemical screening of *Mangifera indica* fruit revealed the presence of sugars, saponins, tannins and flavonoids. Oduro et al. (2009) reported the presence of proximate constituents and phytochemical compounds in two varieties of *T. catappa*, a result which conforms to the findings of the present study.

Table 4. Results of the phytochemical screening of the powdered plant samples

Samples	Anthra-quinines	Alkaloids	Saponins	Tannins	Cardiac glycosides
<i>Mangifera indica</i>	+	+	+	+	+
<i>Terminalia catappa</i>	-	-	+	+	+
<i>Tetracera alnifolia</i>	±	-	+	-	+
<i>Waltheria indica</i>	+	-	+	+	+
<i>Threobroma cacao</i>	+	+	+	+	+
<i>Trema orientalis</i>	-	+	+	+	+
<i>Parquetina nigrescens</i>	-	+	+	+	+
<i>Sorghum bicolor</i>	-	+	±	+	+
<i>Harungana madagascariensis</i>	+	-	+	-	+
<i>Detarium microcarpum</i>	±	+	+	+	+

+ = present; - = absent; ± = inconclusive

4. CONCLUSION

This study, thus, demonstrated that ethnobotanicals aside their therapeutic potential can also function as substitutes for nutritional supplements. *P. nigrescens*, *W. indica* and *T. cacao* could be used as food supplements because of their significant nutritional constituents especially proteins and therapeutic potential. Further search for antianaemic plants should include ethnobotanical survey to preserve indigenous knowledge and upgrade phytomedicines in health care delivery in Nigeria.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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