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Comparative Pharmacognostic Studies of *Vitellaria Paradoxa* in Some Areas of Northern Nigeria

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Abstract

Shea tree (*Vitellaria paradoxa*) is a multipurpose plant utilized by different communities as source income, as food and medicine. The butter produced from the nuts of shea tree have been reported to show variation across various countries with little or no studies on the variation in the other parts of the plant. The shea leaves and stems were collected from Ngaski, Bosso and Yamaltu-Deba local government areas of northern Nigeria and variation in terms of microscopical features and physicochemical properties were studied using standard procedures. The microscopical studies revealed that the plant leaf possesses paracytic type of stomata found only on the abaxial surface with no difference in stomatal number, stomatal index, palisadae ratio, veinslet and vein termination numbers. No difference was also observed on the stem arrangements. The moisture content of leaf from Bosso (LB) was significantly higher than the leaf from Ngaski (LA) and leaf from Yamaltu-Deba (LC), while the moisture content of the stem from Bosso (SB) was significantly lower than the values for stem from Ngaski (SA) and Yamaltu-Deba (SC).There was no significant difference between the total ash values and acid insoluble values for both the leaves and stem, but water soluble ash of SA was significantly lower than SB and SC. Alcohol extractive values of SB was significantly higher than SC but not significantly higher than SA with no significant variation in the water and alcohol extractive values of the leaves. There was significant variation in the moisture contents and extractive values of the plant parts with no significant differences between the microscopic properties of the plant.

Keywords: *Vitellaria paradoxa*, Pharmacognostic, Leaf, Stem, Variation

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INTRODUCTION

Shea tree (*Vitellaria paradoxa*), a monotypic species belonging to the Sapotaceae family is one of the most important agro forest species in Africa occupying a 500 – 700 km wide and 5000 km long stretch of African savanna (Nikiema and Umali, 2002; Maranz*et al*., 2004; Fobil, 2010).The tree is found naturally occupying the wild of the dry savannah belt of West African countries (El-Mahmood *et al*., 2008; Israel, 2015). In Nigeria, the tree grows wild across the guinea and sudan sahel savanna zones covering the entire Northern region including some parts of the South western states (Ololade and Ibrahim, 2014).

The tree is utilized bymany communities and ethnic group in religious and cultural ceremonies as it is considered sacred (Pretarious and Watt, 2001).Traditionally, the leaves, roots, stem, seeds and fruits of shea tree are used in treatment of enteric infection.Decoction of the stem bark is administered to women during delivery to facilitate childbirth and stimulate lactation while the leaves and roots are used in the treatment of stomach pains, nausea, diarrhea, jaundice, epilepsy, convulsion, stress, headache and as eyebath (Ndukwe *et al*., 2005; Bum *et al*., 2011; Usman *et al*., 2014; PROTA, 2019). Research has shown that the root bark possess cytotoxic activity while the stem bark, leaves, fruits and butter were reported to possesses anti-inflammatory, anti-diabetic, anti-microbial and anti-ageing properties (Tapondjou *et al*., 2011; Zhang *et al*., 2014; Eyong *et al*., 2015; Baky *et al*., 2016).

The parts of the plant including thefruits, nuts and leaf have been reported to vary across various agroecological zones in Nigeria. Oyegoke *et al*. (2014) reported that the leaf laminar length, leaf width and petiole length of the shea tree across three locations within northern Nigeria varied significantly. In another study, the fruits from Northern Guinea Savanna of Nigeria were reported to be heavier in weight, while fruits from the Southern Guinea Savanna were fleshier (Ugese *et al*., 2010; Enaberue *et al*., 2014). Ibrahim *et al*. (2020) also reported that the nuts varied in terms of length and width with the shea nuts from Sudan Savannah bigger than those from the Northern Guinea Savanna.

Although, several studies have been carried out on the variations in the shea tree growing in Nigeria, but there are limited studies on the variation of the plant in terms of microscopical and physicochemical parameters, thus the need for the present study.

METHODOLOGY

Collection of Plant Materials

Shea leaves and stems were collected from 20 trees within the tree population between the months of August-September, 2018 from three locations namely; Ngaski (Latitude 11^{o} 14′-12^{o} 39′ North; Longitude 5^{o} 18′-18^{o} 20′), Bosso (Latitude 9^{o} 31′-9^{o} 40′ North; Longitude 6^{o} 29′-6^{o} 35′) and Yamaltu-Deba (Latitude 10^{o} 18′-10^{o} 30′ North; Longitude 11^{o} 20′-11^{o} 40′) local government areas of Kebbi, Niger and Gombe states of Nigeria respectively. The plant materials were transported to the Herbarium unit of the Department of Botany, Ahmadu Bello University, Zaria for proper identification. The leaves and stem of the shea tree were air dried at room temperature, pulverized and stored in airtight container for further use.

Microscopical studies of the shea leaf and stem

The microscopical studies were carried out by clearing the powdered leaves and stems from the study areas, cleared in chloral hydrate with little boiling, the powdered materialwas washed with water. Stained with phloroglucinol and mounted in glycerin for observation. The sections were observed under microscope. The stomatal type, stomatal number, numbers of epidermal cells, palisade ratio, vein islet and vein termination were determined

and the stomatal index calculated. Photograph at different magnification were taken as appropriate (WHO, 2011).

Stomatal Number: The average number of stomata present per square millimeter of the epidermal layer of the leaf.

Stomatal Index: The percentage of the number of stomata to the total number of epidermal cells calculated as follows;

$$\text{Stomatal Index} = \frac{\text{stomatal number}}{\text{epidermal number}} + \text{stomatal number}$$

Palisade ratio: The palisade ratio is the average number of palisade cells that occur beneath an epidermal cell.

$$\text{Palisade ratio} = \frac{\text{Average number of palisade cells beneath one epidermal cell}}{4}$$

Vein-islet Number: The number of vein-islet per square millimeter of the leaf surface.

Vein-let Termination Number: The number of veinlet termination per square millimeter of the leaf surface between the midrib and margin.

Physicochemical studies of the shea leaf and stem

Moisture Contents

Empty crucibles were dried in oven to get a constant weight, five grams (5 g) each of the shea leaves and stem samples were weighed in a dried tared dish and dried in an oven at 105°C. After 1 h, the sample was removed from the oven and placed in the desiccator for 30 min to cool. It was then removed and weighed. This process was repeated until the change of the weight between the two successive observations did not exceed 1 mg (WHO, 2011). The percentage moisture in the samples were calculated as follows;

$$\text{Moisture content (\%)} = \frac{\text{weight of water loss}}{\text{initial wieght of the powdered sample}} \times 100$$

Ash Values

In tared crucibles, 4 g each of the plant materials were weighed and ignited in a furnace at 450°C until white completely. They were cooled in a dessicator and weighed and the ash values were calculated (WHO, 2011).

$$\text{Ash value (\%)} = \frac{\text{weight of residual ash}}{\text{initial weight of powdered sample}} \times 100$$

Acid Insoluble and Water Soluble Ash

The ash obtained from the ash value were boiled in 25 ml of 2N hydrochloric acid for 5 minutes and the insoluble matter collected on ashless filter paper, washed with hot water, ignited and weighed. The percentage of the acid insoluble ash were calculated (WHO, 2011). The ash obtained from the ash value were boiled in 25 ml of water for 5 minutes and the insoluble matter collected on ashless filter papers, washed with hot water, ignited and weighed (WHO, 2011). The percentage of the acid and water insoluble ash were calculated as follows;

$$\text{Water soluble ash values (\%)} = \frac{\text{initial weight of residual ash} - \text{weight of residual ash}}{\text{initial weight of powdered drug}} \times 100$$

$$\text{Acid insoluble ash values (\%)} = \frac{\text{weight of residual ash}}{\text{initial weight of powdered drug}} \times 100$$

Alcohol and Water Extractive Values

The alcohol extractive values were determined by weighing 5 g of the dried powdered plant samples in conical flasks, 100 ml of methanol was added and shaken for 6 hours before allowed to stand for 18 hours (WHO, 2011). The extracts (25 ml) were filtered, evaporated to dryness and the percentage yield calculated. The water extractive values were determined by weighing 5 g of the dried powdered plant samples in conical flasks, 100 ml of distilled water was added and shaken for 6 hours before allowed to stand for 18 hours (WHO, 2011). The extracts (25 ml) were filtered, evaporated to dryness and the percentage yield calculated as follows;

$$\text{Extractive values (\%)} = \frac{\text{weight of extract}}{\text{weight of sample}} \times 100$$

Data Analysis

Analysis of variance (ANOVA) were used to compare the physicochemical variation of the shea leaf and stem by generating the data using SPSS software version 17. Means and standard deviation were computed. Duncan multiple range test were used to compare the mean variance with significance level at $P < 0.05$.

RESULTS

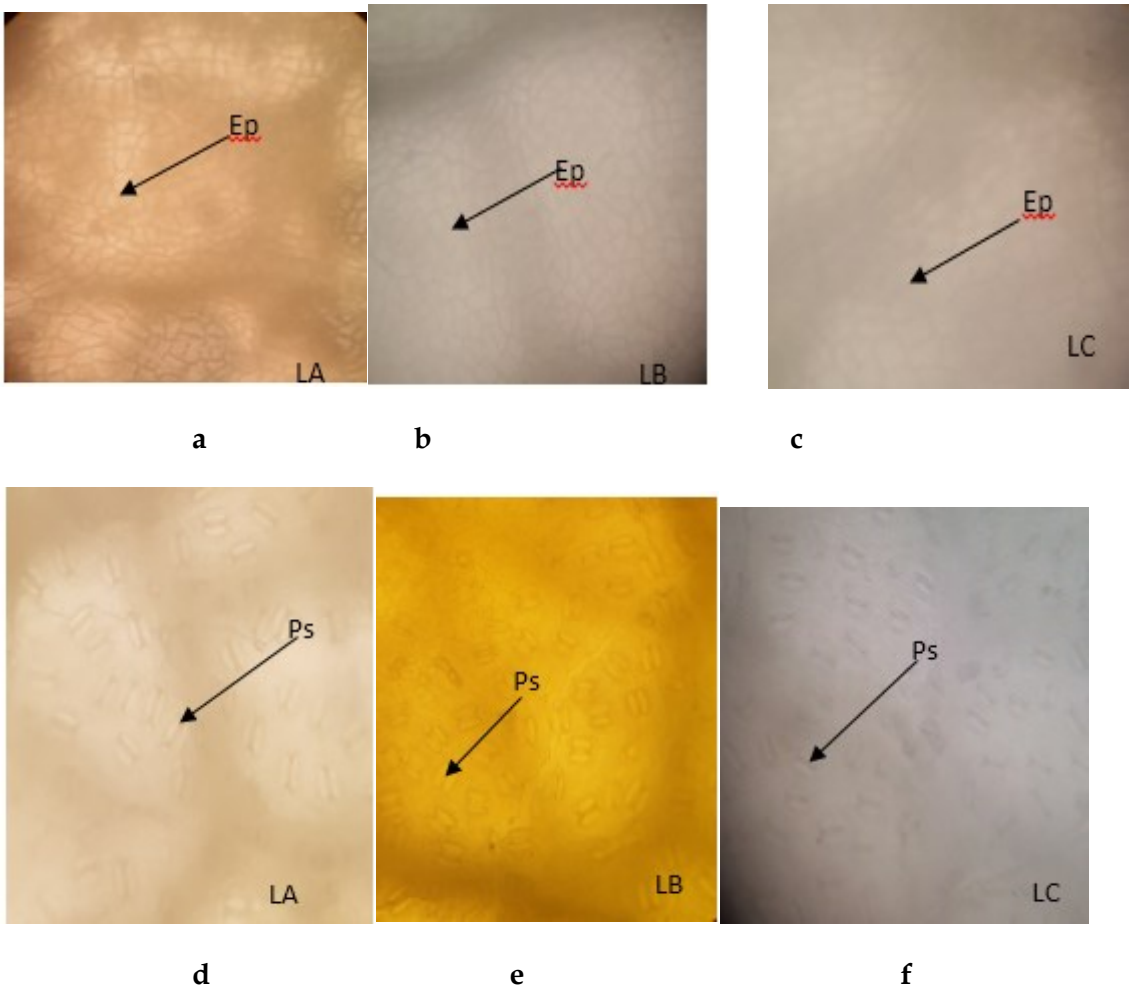
The microscopical studies of leaves of shea tree revealed that the plant is hypostomatic with paracytic stomata (Ps) on the abaxial surfaces (Plates V), while the adaxial surfaces had only epidermal (Ep) cells (Plate V). Vein islet and vein termination were also observed on all the leave surfaces. The stomatal number of the leaves across all the study areas viewed under 20 fields revealed that leaf from Ngaski (LA), leaf from Bosso (LB) and leaf from Yamaltu-Deba (LC) had mean values of stomatal numbers at 27.10 ± 2.51^a, 27.75 ± 2.93^a and 25.833 ± 1.73^a respectively with no significant differences. There was also no significant difference between the stomatal index, palisade ration, vein islet and vein termination numbers of the LA, LB and LC with mean values at 15.71 ± 1.29^a, 16.11 ± 2.10^a and $15.19 \pm 2,12^a$ for stomatal index, 5.37 ± 0.01^a, 5.41 ± 0.01^a and 5.35 ± 0.01^a for palisade ratio, 9.0 ± 0.03^a, 8.96 ± 0.06^a and 9.1 ± 0.14^a for vein islet and 3.30 ± 1.21^a, 3.21 ± 1.70^a and 3.10 ± 1.07^a for the vein termination numbers respectively (Table 1.0).

Microscopical feactures such as collenchyma, sclerenchyma, phloem, xylem, pith, phellogen, phelloderm, pericylicfibres, corks and sclereids were observed on stem from Ngaski (SA), stem from Bosso(SB) and stem from Yamaltu-Deba (SC).

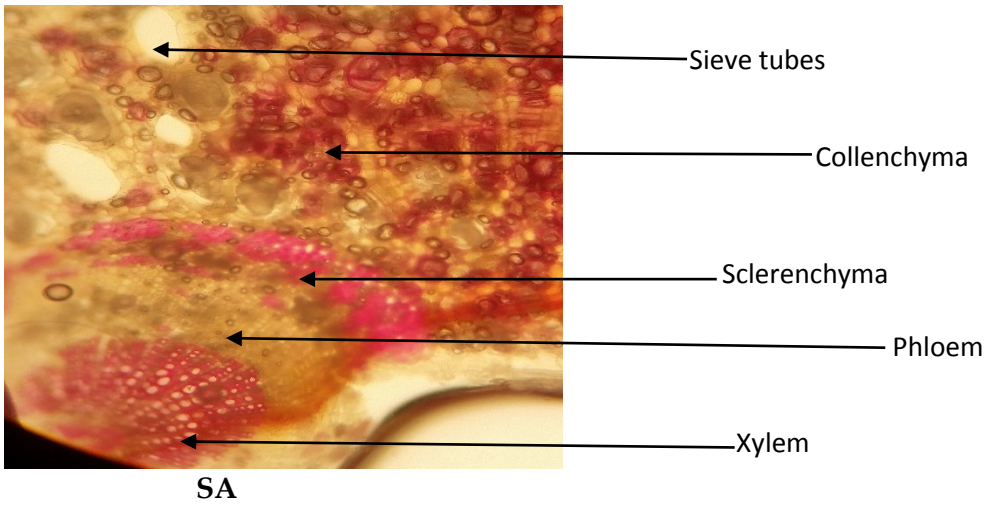
Table 1.0: Quantitative Microscopy of the Shea Leaves

S/No	Parameters	LA	LB	LC
1	**Stomatal number**	27.10 ± 2.51^a	27.75 ± 2.93^a	25.83 ± 1.73^a
2	**Stomatal index**	15.71 ± 1.29^a	15.11 ± 2.10^a	$15.19 \pm 2,12^a$
3	**Palisade ratio**	5.37 ± 0.01^a	5.41 ± 0.01^a	5.35 ± 0.01^a
4	**Vein islet number**	9.00 ± 0.03^a	8.96 ± 0.06^a	9.10 ± 0.14^a
5	**Vein termination number**	3.30 ± 1.21^a	3.21 ± 1.70^a	3.10 ± 1.07^a

Key: Leaves from Ngaski (LA), Leaves from Bosso (LB), Leaves from Yamaltu-Deba (LC)
***SEM with Subscript across row are not significantly different**



Plates I: Showing the (Ep) Epidermal cells on the Upper Surface (a-c) and (Ps) Paracytic Stomatic on the Lower Surface (d-f) on the Shea Leaves from Ngaski (LA), Bosso (LB) and Yamaltu-Deba (LC) at Magnification X10.



SA

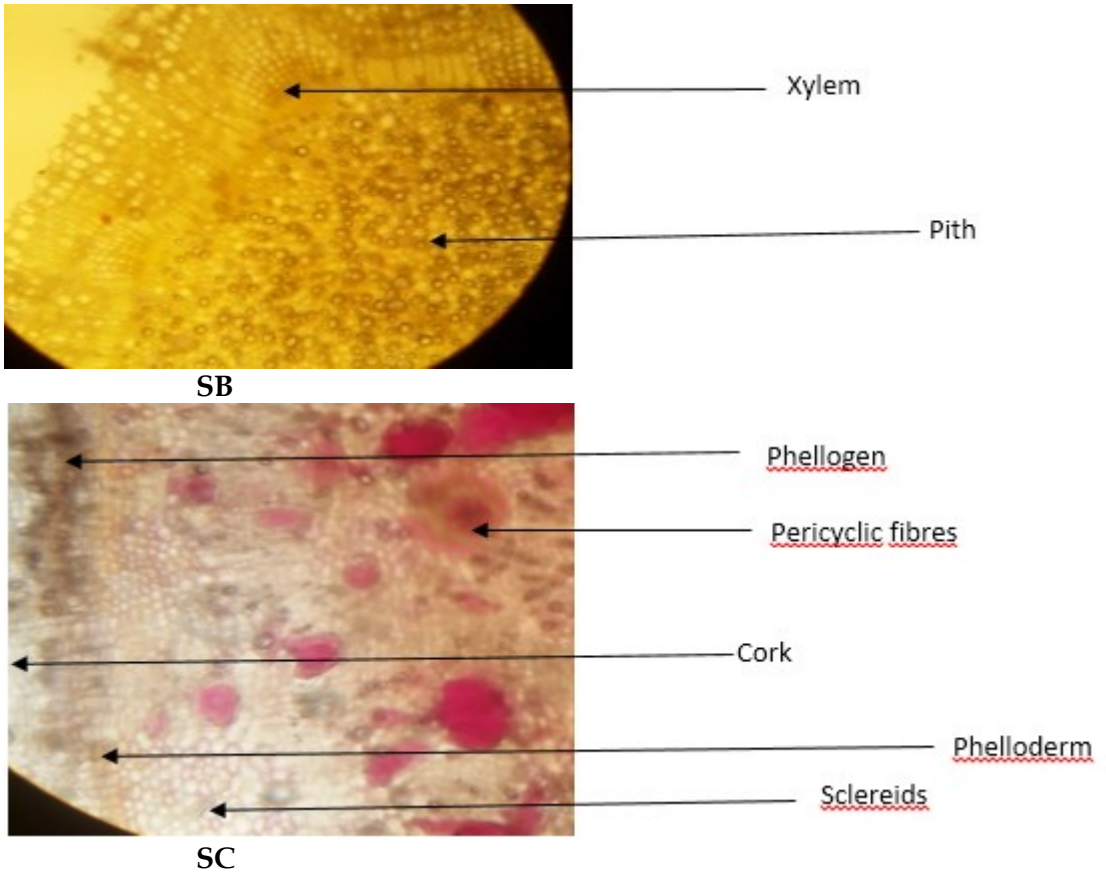


Plate II: Showing the Microscopical Features of the Shea Stem from Ngaski (SA), Bosso (SB) and Yamaltu-Deba (SC) at Magnification X10.

The moisture content of the shea leaves ranged from 1.63 ± 0.2^{a} to 2.86 ± 0.9^{b} %. The shea leaves LA and LC had moisture content at 1.63 ± 0.2^{a} and 2.06 ± 0.2^{a} % respectively which were significantly lower than shea leaves LB at 2.86 ± 0.9^{b} %, while the stem SA and SC at 0.96 ± 0.03^{b} and 0.9 ± 0.1^{b} % respectively which were significantly higher than shea leaves LB at 0.46 ± 0.03^{a} %. There were no significant differences between the ash and acid-insoluble ash values of all the leaves and stem of the shea tree. There was no difference in the water-soluble ash values for the leaves, but SA at 1.69 ± 0.2^{a} % is significantly lower than SB at 3.06 ± 0.7^{b} %, while SC is not significantly different from SA and SB at 2.16 ± 0.4^{ab} %. The water extractive values of the leaves were not significantly different, while SC was significantly higher than SA and SB at 16.7 ± 1.7^{b} %. The alcohol extractive values of LA and LB at 15.0 ± 1.5^{b} % and 16.0 ± 1.2^{b} % were significantly higher than LC at 8.5 ± 0.8^{a} %. The alcohol extractive values of SC at 5.2 ± 1.0^{a} % is significantly lower than SB at 9.93 ± 0.6^{b} %, while there was no significant difference between SA at 7.9 ± 0.6^{ab} % with SB and SC (Table 2.0).

Table 2.0: Physical Parameters of the Shea Leaves and Stem across the Study Areas

S/No.	Samples	Physicochemical Parameters (%) Moisture content	Total ash	Acid insoluble ash	Water soluble ash	Water Extractive values	Extractive Alcohol values
1	LA	1.63±0.20^{a}	3.80±0.90^{a}	1.47±0.30^{a}	2.06±0.10^{a}	18.5±1.80^{a}	15.0±1.53^{b}
2	LB	2.86±0.90^{b}	3.13±1.80^{a}	1.40±0.10^{a}	2.46±1.00^{a}	17.3±1.30^{a}	16.0±1.20^{b}
3	LC	2.00±0.03^{a}	5.80±1.40^{a}	0.90±0.10^{a}	2.86±0.90^{a}	15.3±0.90^{a}	8.5±0.80^{a}
4	SA	0.96±0.03^{b}	3.03±0.60^{a}	1.13±0.60^{a}	1.69±0.20^{a}	16.7±1.7^{b}	7.9±0.86^{ab}
5	SB	0.46±0.03^{a}	5.43±1.50^{a}	1.10±0.60^{a}	3.06±0.70^{b}	11.1±1.50^{a}	9.93±0.60^{b}
6	SC	0.90±0.10^{b}	5.40±1.90^{a}	1.20±0.20^{a}	2.16±0.40^{ab}	10.4±1.13^{a}	5.2±1.00^{a}

Key: Leaves from Ngaski (LA), Leaves from Bosso (LB), Leaves from Yamaltu-Deba (LC), Stem from Ngaski (SA), Stem from Bosso (SB) and Stem from Yamaltu-Deba (SC)
***SEM with same subscript across rows for are not significantly different**

DISCUSSION

Microscopically, the leaves of shea tree across the study areas were found to show no difference in the stomatal numbers, stomatal index, palisade ratio, vein islet number and vein termination number. The stomata type are paracytic occurring only on the upper (abaxial) surface of the leaves, thus they are hypostomatic. This report is in line with the study of Oyegoke *et al* (2014) who studied the shea microscopical features of the shea leaves growing within the north central part of Nigeria and reported absence of trichomes on the leaves and the presence of hypostomatic paracytic stomata. The stem arrangements of the shea tree stem revealed the presence of microscopical features such as the phelloderm, phellogen, sclerenchyma, collenchyma, phloem, xylem and pith are in similar to the microscopical features of *Mimusopselengi* of the same plant family (Kadam *et al*., 2012).

The moisture contents of any given part material provides an idea on its preservation techniques, as the higher the moisture content of any plant material, the more suitable it will provide an enabling environment for microbial growth (Nagani *et al*., 2011). Accordingly, the moisture content of the leaves and stem of the shea trees are within the acceptable limit for the percentage moisture in medicinal plants and as such, can be said to be unfavorable for microbial growth. The total ash, acid-insoluble ash and water soluble ash of the shea leaves and stem are not high, indicating that the plant materials contains low quantity of inorganic and non-physiological matters (Sharma *et al*., 2012). The extractive values of water were found to be higher than methanol for the leaves and stem. This conforms to the claim that water is the most suitable medium for the extraction of bioactive components of (Ajazuddin and Shailendra, 2010).

CONCLUSION

The present study indicated no significant variation in the microscopical and physicochemical parameters of shea trees growing in three geographical areas of northern Nigeria although, there were variations in the moisture content and extractive values which may be as a result of the climatic conditions that vary within the study areas.

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