

ISSN 2411-9547



# ETHIOPIAN JOURNAL OF BIODIVERSITY (EthJBD)

**Official Journal of the  
Ethiopian Biodiversity Institute**

April, 2023

Addis Ababa, Ethiopia

**VOLUME 4  
No. 1**

## **Ethiopian Journal of Biodiversity (EthJBD)**

The Ethiopian Journal of Biodiversity (EthJBD) is a journal published biannually by the Ethiopian Biodiversity Institute (EBI). EthJBD publishes peer reviewed scientific articles in areas of biodiversity, agriculture, natural resource use, community knowledge, access and benefit sharing, environment, climate change, modeling and related areas.

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ETHIOPIAN JOURNAL OF BIODIVERSITY

VOLUME 4, NO. 1 (APRIL 2023)

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**ASSESSMENT OF ETHIOPIAN BARLEY (*HORDEUM VULGARE L.*) GENOTYPES  
RESISTANCE FOR SCALD (*RHYNCHOSPORIUM COMMUNE*) AND POWDERY  
MILDEW (*BLUMERIA GRAMINIS*)**

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**ABSTRACT:** The existence of a high degree of variability in crop genetic resources for disease resistance can be taken as one of the strategies that allow crops to survive in equilibrium with the challenges posed by pathogenic organisms. Three hundred twenty barley (*Hordeum vulgare L.*) lines were evaluated for severity and area under disease progress curve (AUDPC) at two locations in Ethiopia. These disease resistance traits were for barley leaf scald (*Rhynchosporium commune*) and powdery mildew (*Blumeria graminis*). The lines were tested during the 2017/18 and 2018/19 cropping seasons for disease severity and in a single season for AUDPC, under natural infestation conditions. The REML analysis based on BLUP mean revealed that lines differ significantly ( $p \leq 0.01$ ) for both disease severity and AUDPC in both diseases. Furthermore, there was a significant difference ( $p \leq 0.01$ ) among the test environments for disease severity based on over years combined data. For leaf scald, the severity was found to be higher in the first year possibly due to a comparatively earlier planting date in the season which in turn facilitated repeated infection within the growing season. Hence, apart from the utilization of resistant lines, avoiding early planting can be taken as a good management strategy to reduce the damage caused by leaf scald. Lines HB-42 and Accn# 243209 were in the top ten for both leaf scald and powdery mildew resistance suggesting their potential for multiple disease resistance. These two lines can be used as parental lines in the attempt to develop high-yielding genotypes with multiple disease-resistance backgrounds.

**Keywords:** AUDPC, disease severity, genetic resources, farmers' varieties.

## INTRODUCTION

In this era of climate change crop production can be hampered by various biotic and abiotic factors. In any breeding program to develop varieties of a crop of interest that can exist in equilibrium with challenges caused by pathogenic organisms, the availability of an important source of variability is imperative (Zhao et al., 2022). Reduction in the genetic base of crops, on the contrary, increases crop vulnerability to various pathogens (Bailey-Serres et al., 2019; Zhao et al., 2022).

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Concerning barley production in the world, diseases caused by fungal pathogens represent the key constraint, despite substantial efforts to manage the damage. Cultivated barley hosts more than 250 different pathogens with variable levels of importance (Walters et al., 2012; Singh et al., 2019). Repeated resistance breakdown in host plants as a result of increased pathogenicity is causing a threat to global barley production. Leaf scald, caused by *Rhynchosporium commune* (Avrova and Knogge, 2012), and powdery mildew, caused by *Blumeria graminis* f. *hordie*, are among the main and most widely distributed diseases in barley (Spies et al., 2012).

Previously, *Rhynchosporium secalis* was known to cause leaf blotch in rye, triticale and barley. However, with advances in pathogenicity studies, it was later discovered this pathogen has further host specialization over the mentioned crops. Hence, the pathogen causing scald in barley was reclassified to be *Rhynchosporium commune*, whereas the pathogen causing leaf blotch in rye and triticale remained *Rhynchosporium secalis* (Zaffarano et al., 2011; Avrova and Knogge, 2012; King et al., 2015; Zhang et al., 2020). Barley leaf scald is a haploid, polycyclic fungus with repeated pathogen generations in one growing season (Avrova and Knogge, 2012). The primary inoculum is coming either from crop debris or infected seed. Splash-dispersal from leaves infected by the pathogen are the source of secondary disease spread (Davis and Fitt, 1992; Zhan et al., 2008). *Rhynchosporium commune* a hemibiotrophic fungus (Oliver and Ipcho, 2004; Zhan et al., 2008) that can cause yield loss from 15 to 45% and reduce grain quality (Brown, 1985); 30 to 40% (Paulitz and Steffenson, 2010). In Ethiopia, it is the most widely distributed and destructive disease of barley. The disease is most prevalent in the highlands characterized by high rainfall and low temperature during the cropping season (Meles et al., 2004).

Powdery mildew, an ascomycetes fungi affects more than 10,000 plant species (Takamatsu, 2004; Kusch et al., 2023). *Blumeria graminis* f. sp. *Hordei* (*Bgh*) is a wind-borne pathogen that relies entirely on its host for its growth and reproduction as is an obligate biotroph (Both et al., 2005; Rsaliyev et al., 2017; Piechota et al., 2019). *Bgh* is the most widespread barley pathogen worldwide and causes yield reduction of 5-10%

and in severe cases yield loss may reach up to 40% (Chaure et al., 2000; Piechota et al., 2019). Owing to the potential to produce large numbers of sexual haploid spores, sexual recombination during the growing season and airborne dispersal over large distances, *Bgh* has a very rapid evolution. This in turn resulted in the development of stable resistance to be very difficult (Bouguennec et al., 2016).

Eighteen different genes conferring resistance to *R. commune* have been reported in barley, many of which have been mapped. On chromosome 3H more than 11 alleles were described (Bjørnstad et al., 2002), *Rsl5b* on chromosome 2H (Schweizer et al., 1995), *Rrs14* on chromosome 1H (Garvin et al., 2000), *Rrs16* on chromosome 4H (Pickering et al., 2006), *Rrs13* on chromosome 6H (Abbott et al., 1995), *Rrs2, Rsl5a* on chromosome 7H (Genger et al., 2005; Hanemann et al., 2009). In addition, several QTL for scald resistance were identified on all chromosomes but 5H (Wang et al., 2014). Barley resistance genes known to confer resistance against powdery mildew could be race-specific (more than 85) (Jørgensen, 1994) or race-non-specific. The powdery mildew resistance genes mapped on the barley genome include *Mla*, the most thoroughly characterized race-specific locus conferring resistance to powdery mildew, *Mlat*, *MlGa*, *Mlk*, *Mlnn*, and *Mlra* on chromosome 1H; *MILa* on 2H; *mlo*, *Mlg*, and *MIBo* on 4H; *Mlj* on 5H; *Mlh* on 6H; and *mlt* and *Mlf* on 7H (Jørgensen, 1994; Schönfeld et al., 1996).

Management of diseases following cultural methods is considered environmentally friendly, but is often less effective, especially under high disease incidence situations. Chemical control methods in Ethiopia are the last option because of the associated higher cost and their adverse effect on the environment. For this reason, genetic resistance is the best option for the sustainable management of pathogens. Deployment of resistant varieties is the most economically effective and environmentally friendly way to cope with the damage caused by pathogens. *mlo*, a recessive allele identified from an Ethiopian barley farmer variety, was found to be a very effective and durable source of resistance against barley powdery mildew. As a result, this resistance gene has been incorporated to a larger extent in European barley varieties (Piechota et al., 2019). Farmers' varieties and breeding materials resistant to scald were also identified (Daba et al., 2019).

However, the ability of the pathogens to evolve rapidly and generate new virulent pathotypes in a short period of time emphasizes the significance of hunting for new sources of resistance genes. To this end, farmers' variety collections represent valuable reservoirs of genetic diversity, which have not been fully employed, and could be successfully exploited in modern breeding programs for disease resistance (Jørgensen and Jensen, 1997). The study aimed to assess Ethiopian barley farmers' and improved varieties' resistance to leaf scald and powdery mildew diseases.

## **MATERIALS AND METHODS**

### **Genetic materials**

The current study comprises 320 barley lines where, 249 barley accessions originally obtained from the Ethiopian Biodiversity Institute were used to develop 501 lines through spike-to-row maintenance, of which 293 were included in this experiment. In addition to the developed lines, 27 varieties developed, released and maintained by national and regional agricultural centers were included, bringing the total number to 320. The experimental materials comprised 6-rowed, 2-rowed (both deficient and male fertile) and irregular (labile) barley variants.

### **Field experiment**

The experiment was laid out in two replications in four rows of 2.5m length. The size of each plot was 2m<sup>2</sup> and 17g of seed was sown to each of the plots. The experiment was conducted at two locations (Arsi Negelle and Holeta) for two years though the type of disease over the location and type of disease-specific traits over the years were variable. The plots were fertilized with DAP and Urea fertilizers as per the recommended rate of applications for the two sites and other agronomic management were uniformly applied to all the lines.

### **Data collection**

The disease data were collected from lines in which the pathogens grew under natural infestation conditions. Scald disease severity was scored visually on plot bases (Amezrou et al., 2018) following a double-digit

scale (D1D2, 00-99) in the experimental fields. The first digit (D1) indicates vertical disease progress on the plant and the second digit (D2) refers to severity measured as diseased leaf area (Saari and Prescott, 1975; Eyal et al., 1987). The disease severity percentage was computed using the following formula (Sharma et al., 2007).

$$\text{Disease severity}\% = \left(\frac{D1}{9}\right) \left(\frac{D2}{9}\right) \times 100.$$

Powdery mildew severity was assessed visually as well, however, scored as a single digit (0-9) scoring scale (Saari and Prescott, 1975) and hence disease severity percentage was estimated as follows.

$$\text{Disease severity \%} = \left(\frac{D}{9}\right) \times 100$$

where D is the disease severity measured as a progress over the height of the plant and diseased leaf area. For both of the diseases scoring was done two times in the first year and four times in fifteen-day intervals in the second year and the final score was taken for percentage disease severity estimation. According to the severity percentage lines can be grouped as highly resistant (0–5%), resistant (5–10%), moderately resistant (10–20%), moderately susceptible (20–30%), susceptible (30–40%), and highly susceptible (> 40%) (Eyal et al., 1987). The disease severity percentage values were Arc-sine transformed.

The area under the disease progress curve (AUDPC) (percent days) gives a quantitative measure of epidemic development and disease intensity (Das et al., 1992). It was computed based on the transformed disease severity percentage corresponding to the four records using the following formula:

$$\text{AUDPC} = \sum_{i=1}^{n-1} \left[ \frac{(X_i + X_{i+1})}{2} \right] (T_{i+1} - T_i)$$

where  $X_i$  = the disease severity on the  $i$ th date,  $T_{(i+1)} - T_i$  = time or days between two disease scores,  $n$  = number of dates on which the disease was recorded. The disease data collected varied depending on the locations and year. Scald severity was estimated in years (2017/18 and 2018/19) at Holeta and scald AUDPC was estimated only in the second year (2018/19) at Holeta. Disease severity for powdery mildew was scored in both years and AUDPC was estimated only in 2018/19 at Arsi Negelle. Lower AUDPC values are associated



with a better resistance of a particular line to a particular disease and a higher AUDPC score means higher susceptibility.

### Statistical analysis

The restricted maximum likelihood (REML) algorithm was used to produce the best linear unbiased prediction (BLUP) mean, in which lines and environments were fitted as random effects. Both scald and powdery mildew severity BLUP mean was combined over two seasons. BLUP means for scald and powdery mildew AUDPC were estimated from a single environment. It was calculated using the META- R statistical software version 6.04 (Alvarado et al., 2019). The model used to generate the combined BLUP means was as follows:

$$Y_{ijk} = \mu + \text{Env}_i + \text{Rep}_j(\text{Env}_i) + \text{Block}_k(\text{Env}_i\text{Rep}_j) + \text{Gen}_l + \text{Env}_i \times \text{Gen}_l + \epsilon_{ijkl}$$

where  $Y_{ijk}$  is the trait of interest,  $\mu$  is the general mean,  $\text{Env}_i$  is the effect of the  $i$ th environment,  $\text{Rep}_j(\text{Env}_i)$  is the effect of the  $j$ th replicate within the  $i$ th environment,  $\text{Block}_k(\text{Env}_i\text{Rep}_j)$  is the effect of the  $k$ th incomplete block within the  $i$ th environment and  $j$ th replicate,  $\text{Gen}_l$  is the effect of the  $l$ th genotype,  $\text{Env}_i \times \text{Gen}_l$  is genotype by environment interaction and  $\epsilon_{ijkl}$  is the error associated with the  $i$ th environment,  $j$ th replication,  $k$ th incomplete block and the  $l$ th genotype, which is assumed to be normally distributed. On the other hand, the model used to produce individual location BLUP mean was:

$$Y_{ijk} = \mu + \text{Rep}_i + \text{Block}_j(\text{Rep}_i) + \text{Gen}_k + \epsilon_{ijk}$$

where  $Y_{ijk}$  is the trait of interest,  $\mu$  is the mean effect,  $\text{Rep}_i$  is the effect of the  $i$ th replicate,  $\text{Block}_j(\text{Rep}_i)$  is the effect of the  $j$ th incomplete block within the  $i$ th replicate,  $\text{Gen}_k$  is the effect of the  $k$ th genotype,  $\epsilon_{ijk}$  is the error associated with the  $i$ th replication,  $j$ th incomplete block and the  $k$ th genotype. The box plot and correlation among variables were performed using the ggplot 2 function of R statistical software version 3.6.1(R core team, 2019).

## RESULTS

The panel of barley lines was evaluated for scald and powdery mildew depending on the prevalence of the diseases at Arsi Negelle and Holeta respectively under natural infestation. The lines showed highly significant ( $p \leq 0.01$ ) variation for both disease severity and AUDPC. Treatment by environment (year) interaction effects for disease severity was also highly significant ( $p \leq 0.01$ ) for both leaf scald and powdery mildew, suggesting a differential response of the genotypes over the test years (Table 1; Table 2).

**Table 1.** REML variance component analysis for barley leaf scald based on individual and combined environments for severity and AUDPC disease traits.

Source of variation	Year			Combined over years Scald severity (%)
	2017/18 Scald severity	2018/19 Scald severity	Scald AUDPC	
Year				199.68**
Genotype	418.35**	191.92**	134960.3**	490.09**
Genotype x year				122.54**
Residual	22.85	19.77	19850.66	21.62
Grand Mean	41.21	31.76	919.38	36.48
LSD	6.65	5.85	187.60	7.88
CV (%)	11.60	13.99	15.32	12.74

AUDPC= Area under disease progress curve, \*\*=highly significant, \*=significant

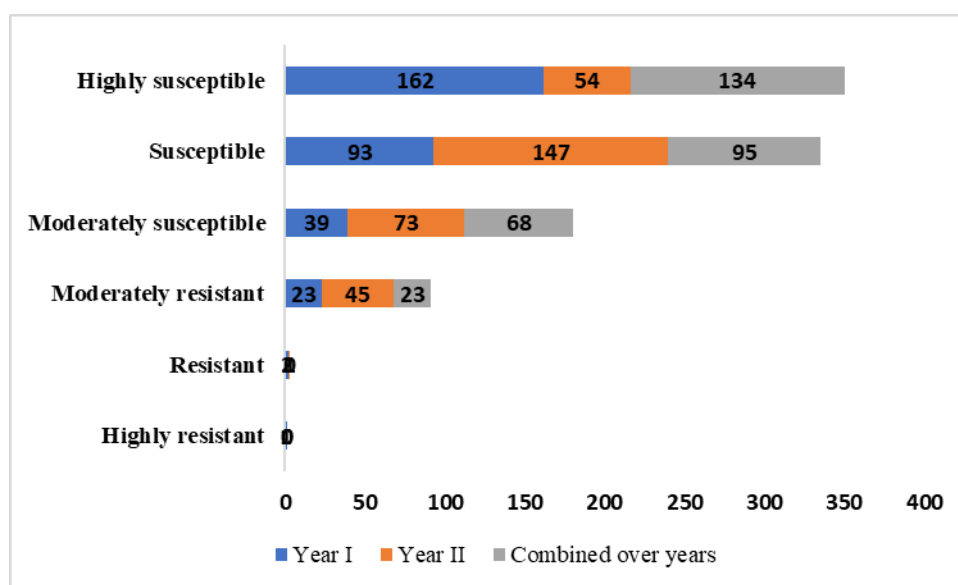
**Table 2.** REML variance component analysis for barley powdery mildew based on individual and combined environments for severity and AUDPC disease traits.

Source of variation	Year			Combined over years Powdery mildew severity (%)
	2017/18 Powdery mildew severity (%)	2018/19 Powdery mildew severity (%)	Powdery mildew AUDPC	
Year				38.82 <sup>ns</sup>
Genotype	204.86**	260.85**	323558**	371.02**
Genotype x year				92.86**
Residual	53.25	58.56	42065.55	55.97
Grand Mean	53.58	52.52	1852.80	53.05
LSD	8.88	9.37	269.68	7.72
CV (%)	13.61	14.56	11.07	14.10

AUDPC= Area under disease progress curve, ns=non-significant, \*\*=highly significant

## Leaf scald

Although there was seasonal variation, it was possible to classify the barley lines from highly resistant to highly susceptible to leaf scald. In 2017/18, only variety HB-42 was found to be highly resistant (Figure 1; Table 3). Two lines, Acc# 24638-B, Acc# 16866 (0.6%), were resistant, 23 lines (7.1%) were moderately resistant, 39 lines (12.1%) turned out to be moderately susceptible and 93 lines (29%) were classified as susceptible; fifty percent of the lines fell in the highly susceptible category. In the second year (2018/19) on the other hand none of the lines appeared in the highly resistant groups. In this case, the majority of the lines were found to be susceptible but there was one variety (EH-1847) with resistant response and 45 lines (14%) with moderately resistant response. Combined over the two years, the majority of the lines were in the highly susceptible group for this disease. No lines were identified in the highly resistant or the resistant group, however, 23 lines (7.1%) were classified as moderately resistant.



**Figure 1.** The distribution of the barley lines for leaf scald severity in each of the severity groups over the individual years and combined over the two years. The X-axis is the number of barley genotypes in each severity group and the y-axis severity groups for leaf scald.

**Table 3.** The response of the top 25 barley lines to severity and AUDPC disease traits of leaf scald on individual years and combined over the test years.

Serial no.	First year (2017-2018)		Second year (2018-2019)		Combined over years		Second year (2018-2019)	
	Genotype	Severity	Genotype	Severity	Genotype	Severity	Genotype	AUDPC
1	243209-A	20.42	HB-42	11.79	Bahati	14.02	Bahati	233.97
2	Bahati	21.21	Explorer	11.79	HB-42	14.29	HB-42	249.83
3	Traveller	21.29	Traveller	11.79	Traveller	14.31	Traveller	250.39
4	HB-42	21.58	Bahati	11.79	243209-A	17.64	Explorer	255.22
5	219026-B	21.95	64334	19.34	219026-B	18.48	243209-A	425.35
6	238360	25.90	219026-B	19.34	Explorer	18.66	219026-B	428.86
7	A hore 880/61	28.11	238360	19.34	238360	20.41	238360	439.97
8	208842-A	28.56	243209-A	19.34	208842-A	25.54	64334	448.70
9	Explorer	29.33	64333-B	22.70	64333-B	25.99	Derebie	676.83
10	64333-B	32.71	208842-A	26.89	A hore 880/61	26.10	64333-B	716.42
11	208841-A	37.87	A hore 880/61	26.89	64334	31.63	HB 1964	743.47
12	243307-A	38.56	HB 1964	26.89	HB 1964	31.86	A hore 880/61	771.20
13	Ibon174/03	39.39	Derebie	35.78	222969-A	38.06	HB-1965	821.44
14	HB 1964	39.57	HB-1965	35.78	Ibon174/03	40.82	208842-A	872.23
15	222969-A	39.73	17651	39.14	208841-B	41.86	Ibon174/03	1051.63
16	213594-A	42.42	18318-B	39.14	213594-A	42.76	17658	1296.15
17	219580-A	44.85	208841-B	39.14	Holker	42.82	219580-A	1318.89
18	17244-A	44.97	222969-A	39.14	208841-A	43.10	17663	1387.85
19	64334	45.68	242093-A	39.14	17244-A	43.47	HB-1963	1412.67
20	208841-B	46.15	Fanaka	39.14	243307-A	43.74	3514-C	1412.69
21	219612-A	47.25	Holker	39.14	18318-B	43.78	64336-A	1418.28
22	204802-B	47.27	17244-A	44.15	242093-A	43.82	213527-A	1424.50
23	Bekoji-1	47.46	17252-C	44.15	Derebie	43.91	4540-A	1429.07
24	Holker	47.50	17658	44.15	Fanaka	43.98	243307-A	1442.06
25	208816-A	47.53	17663	44.15	17651	44.36	222969-A	1499.58

The barley lines showed a considerably wider range for both severity and AUDPC for leaf scald (Table 4a).

In 2017/18, the barley lines varied for leaf scald from 1.89 % (HB-42) to 62.24% (Accession# 237329). In 2018/19, it ranged from 9.55% (EH-1847) to 51.26 (Accession # 208923, 213527-A, 215217-A, 219026-B, 221325, 230814-A). Comparing the individual years, the range for scald severity was wider in the first year than in the second year (Figure 2). Besides, the overall mean performance of the lines for leaf scald severity was higher in the first year than in the second year. In accordance with this, the number of lines classified as susceptible (from moderately susceptible to highly susceptible category) was lower in the second year than the first, because of the higher disease pressure in the first year. Out of the 23 lines that were in the

moderately resistant category combined over the test years, 13 (57%) were lines derived from farmer's varieties and 10 were improved varieties (Table 3). The variety HB-42 was found to be the most resistant one (11.62%) and the lines from farmers' varieties Accession# 208923, 213527-A and 221325 were the most susceptible with a severity percentage of 55%. Leaf scald AUDPC on the other hand varied between 342.46 (EH-1847) to 1584.95 percent days (Accession# 221325). In the best ten lines characterized by a slow scald development (AUDPC), 5 were improved varieties and among these two, HB-42 and EH-1847 were also in the top ten least scalding (lower scald severity). Likewise, five lines from farmers' varieties, i.e. Accession# 243209-A, 16866, 17148, 24639-A, 242093-A, with slow scald development were also in the top ten least scalding groups. In response to leaf scald, the lines of our panel showed a skewed distribution towards high susceptibility for both severity and AUDPC disease traits (Appendix 1). This pseudo-normal distribution may indicate the quantitative nature of both leaf scald resistance within the lines and varieties.

**Table 4.** The range of the mean performance of 320 barley lines for severity and AUDPC of leaf scald and powdery mildew based on individual years and combined over years.

a) Leaf scald

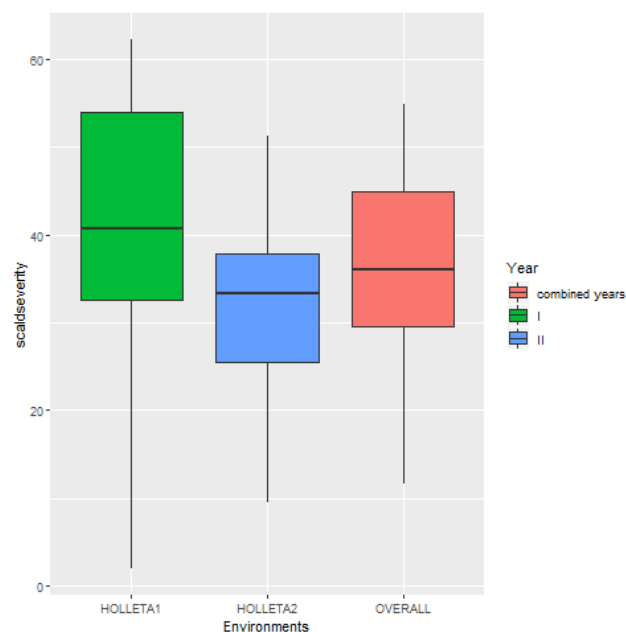
Traits	Year					
	2017/18		2018/19		Combined over years	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Severity	1.89	62.24	9.55	51.2	11.63	54.92
AUDPC			342.46	1584.95		

AUDPC= Area under disease progress curve

b) Powdery mildew

Traits	Year					
	2018		2019		Combined over years	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Severity	20.42	79.83	11.79	67.91	14.02	69.65
AUDPC			233.97	2353.34		

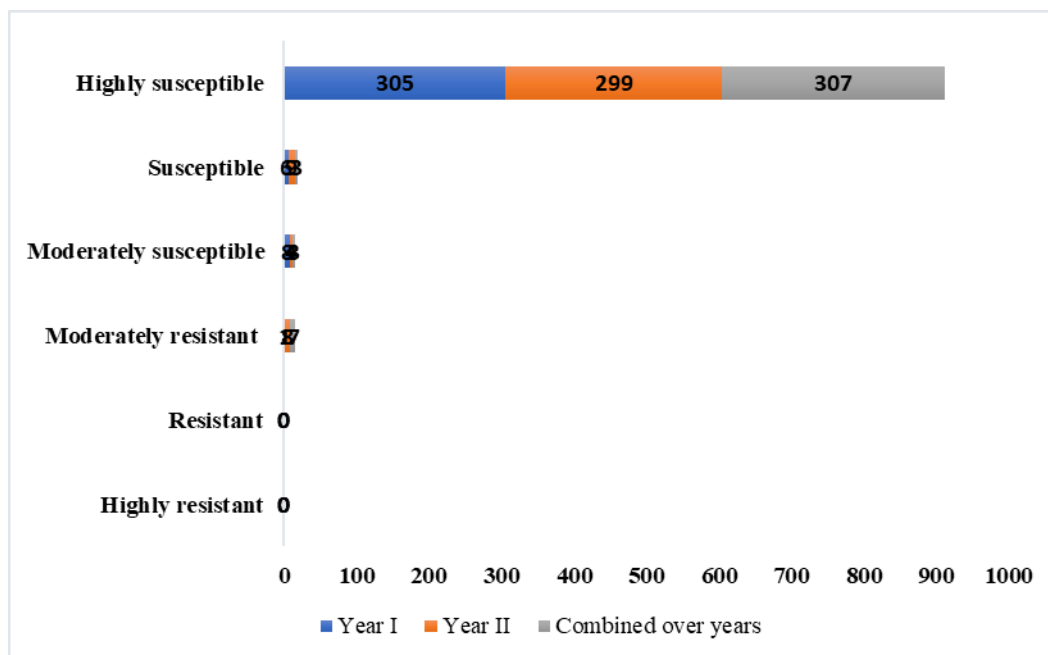
AUDPC= Area under disease progress curve



**Figure 2.** Leaf scald severity of the barley lines across the evaluation years. The x-axis shows the test environments (HOLLETA1=Holeta first year, HOLLETA2=Holeta second year and OVERALL=combined over the years) indicated as per the legend. The y-axis indicates the leaf scald severity level.

#### Powdery mildew

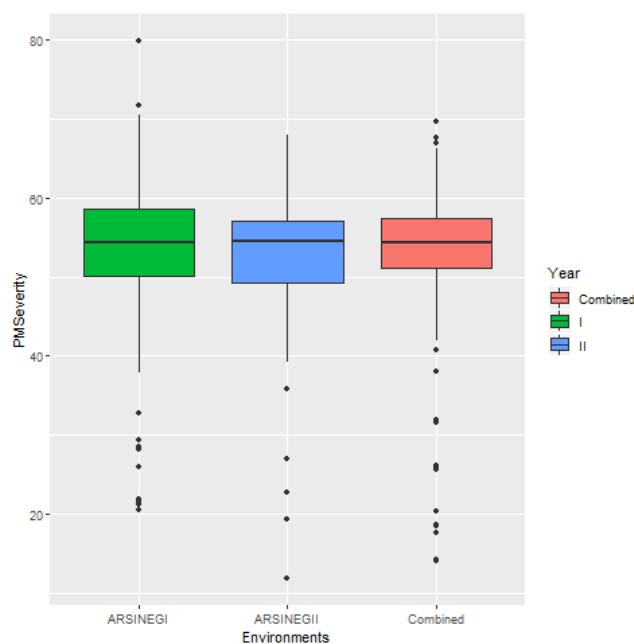
The evaluation of the performance of the barley lines to powdery mildew revealed that the majority were highly affected by the disease. In the first year, no single line was recorded to be either highly resistant or resistant, only one line was moderately resistant (243209-A) (Figure 3). In the second year, although no line was either highly resistant or resistant, eight lines (2.5%) were moderately resistant. Combined over the years seven lines (2.2%) were found to be moderately resistant whereas the rest were in the susceptibility category.



**Figure 3.** The distribution of the barley lines for powdery mildew severity to each of the severity groups over the individual years and combined over the two years. The X-axis is the number of barley genotypes in each severity group and the y-axis severity groups for leaf scald.

The barley lines also showed a wide range for both disease traits (severity and AUDPC) (Table 4b). For powdery mildew severity in the first year, the lines varied from 20.42% (Accession# 243209-A) to 79.83% (Accession#17244-B). In the second year, it spanned between 11.79% (HB-42) to 67.91% (Accession# 3545-C). The number of lines in the moderately resistant category increased from one in the first year to eight in the second year. Although there was no considerable variation in the range over the two years, the minimum and the maximum values in the second year were lower than it was in the first year (Figure 4) and had comparable mean performance over the two years. Seven barley lines (2.18%) based on over years combined data were found to be moderately resistant to barley powdery mildew and out of these lines four (57%) were improved varieties and three were lines from farmers' varieties. The line with the lowest severity percentage for powdery mildew was the improved variety Bahati (14.01%) (Table 5) and the most susceptible variety with a severity percentage of 69.65% was Accession# 17244-B. The same variety (Bahati) with 233.97 percent days showed the lowest AUDPC value and the line from farmer variety

Accession # 64233-C with 2353.34 percent days had the highest AUDPC. Five of the ten lines with low AUDPC were derived from farmers' varieties and were also in the top ten lines with low disease severity. Out of the five varieties with the lowest AUDPC all but one variety (Derebie) were in the top ten lines with low severity. For both severity and AUDPC disease traits the mean performance of the lines followed a skewed distribution towards higher susceptibility for both of the test years and combined over the years (Appendix 2).



**Figure 4.** Leaf scald severity of the barley lines across the evaluation years. The x-axis shows the test environments (ARSINEGI= Arsi Negelle year I, ARSINEGII= Arsi Negelle year II and combined= combined over the two years) indicated as per the legend. The y-axis indicates barley powdery mildew severity level.



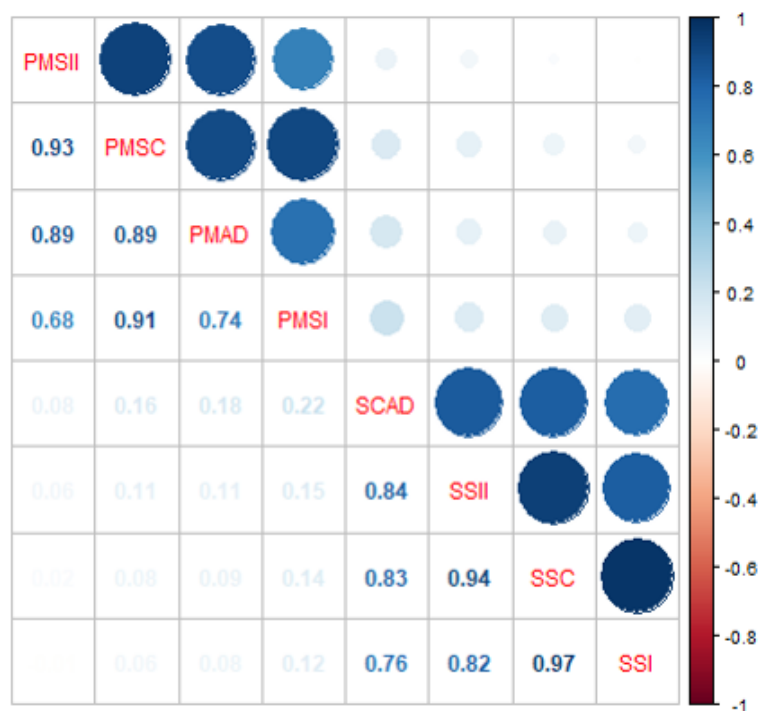
**Table 5.** The response of the top 25 barley lines to severity and AUDPC disease traits of powdery mildew on individual year and combined over the test year.

Serial no.	First year (2017-2018)		Second year (2018/19)		Combined over years		Second year (2018-2019)	
	Genotype	Severity	Genotype	Severity	Genotype	Severity	Genotype	AUDPC
1	243209-A	20.42	HB-42	11.79	Bahati	14.02	Bahati	233.97
2	Bahati	21.21	Explorer	11.79	HB-42	14.29	HB-42	249.83
3	Traveller	21.29	Traveller	11.79	Traveller	14.31	Traveller	250.39
4	HB-42	21.58	Bahati	11.79	243209-A	17.64	Explorer	255.22
5	219026-B	21.95	64334	19.34	219026-B	18.48	243209-A	425.35
6	238360	25.90	219026-B	19.34	Explorer	18.66	219026-B	428.86
7	A hore 880/61	28.11	238360	19.34	238360	20.41	238360	439.97
8	208842-A	28.56	243209-A	19.34	208842-A	25.54	64334	448.70
9	Explorer	29.33	64333-B	22.70	64333-B	25.99	Derebie	676.83
10	64333-B	32.71	208842-A	26.89	A hore 880/61	26.10	64333-B	716.42
11	208841-A	37.87	A hore 880/61	26.89	64334	31.63	HB 1964	743.47
12	243307-A	38.56	HB 1964	26.89	HB 1964	31.86	A hore	771.20
13	Ibon174/03	39.39	Derebie	35.78	222969-A	38.06	HB-1965	821.44
14	HB 1964	39.57	HB-1965	35.78	Ibon174/03	40.82	208842-A	872.23
15	222969-A	39.73	17651	39.14	208841-B	41.86	Ibon174/03	1051.63
16	213594-A	42.42	18318-B	39.14	213594-A	42.76	17658	1296.15
17	219580-A	44.85	208841-B	39.14	Holker	42.82	219580-A	1318.89
18	17244-A	44.97	222969-A	39.14	208841-A	43.10	17663	1387.85
19	64334	45.68	242093-A	39.14	17244-A	43.47	HB-1963	1412.67
20	208841-B	46.15	Fanaka	39.14	243307-A	43.74	3514-C	1412.69
21	219612-A	47.25	Holker	39.14	18318-B	43.78	64336-A	1418.28
22	204802-B	47.27	17244-A	44.15	242093-A	43.82	213527-A	1424.50
23	Bekoji-1	47.46	17252-C	44.15	Derebie	43.91	4540-A	1429.07
24	Holker	47.50	17658	44.15	Fanaka	43.98	243307-A	1442.06
25	208816-A	47.53	17663	44.15	17651	44.36	222969-A	1499.58

### Correlation among leaf scald and powdery mildew disease traits

In order to determine the degree of association among the diseases, the disease traits and the test environments, the BLUP mean of the response of the lines for severity and AUDPC traits of both barley leaf scald and powdery mildew were analyzed for correlations (Figure 5). Correlation coefficients were calculated among disease traits within and between the diseases, among the test environments for both diseases. Significant, and positive correlations were observed between severity and AUDPC disease traits for both leaf scald and powdery mildew ( $r=0.83$  between combined leaf scald severity and AUDPC,  $r=0.89$

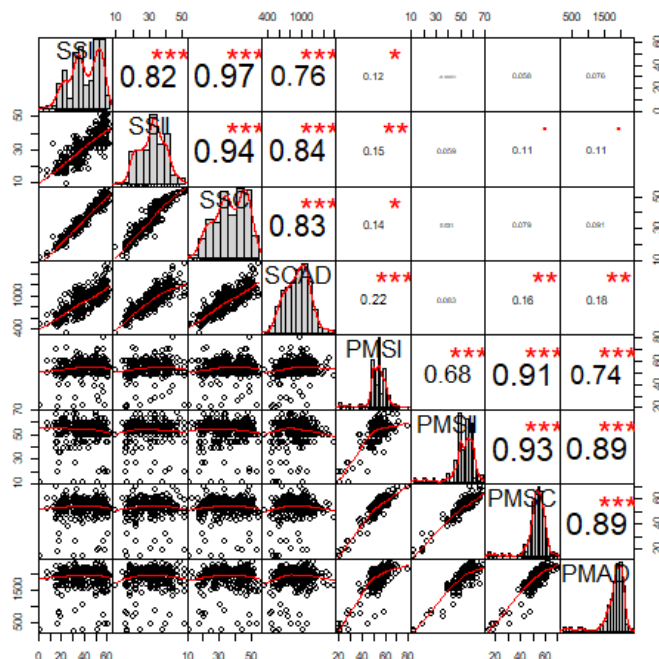
between combined powdery mildew severity and AUDPC). Likewise, correlations between the different test environments were also significantly high and positive ( $r=0.82$  between severity in years I and II for scald and  $r=0.68$  between severity in years I and II for powdery mildew). Differently, correlations of disease traits between the two diseases were found to be weak though positive ( $r=0.078$  between leaf scald severity combined over years and powdery mildew severity combined over years).



**Figure 5.** Correlation coefficients for leaf scald and powdery mildew disease traits SSI= leaf scald severity year one, SSII= leaf scald severity year II, SSC= leaf scald severity combined over years, SCAD= leaf scald area under disease progress curve, PMSI= powdery mildew severity year one, PMSII= powdery mildew severity year two, PMSC= powdery mildew severity combined over the test years, PMAD= powdery mildew area under disease progress curve. The strength and direction of the correlation are represented by the color and size of the circle in relation to the legend.

Correlation between the disease traits and test environments was highly significant ( $P \leq 0.01$ ) (Figure 6) for both leaf scald and powdery mildew. The high, positive and significant correlation between severity combined over years and AUDPC  $r=0.84$  for leaf scald and  $r=0.89$  for powdery mildew suggests the lower

the severity percentage, the lower the AUDPC and hence the better the response of the line to the particular disease.



**Figure 6.** Correlation coefficients for leaf scald and powdery mildew disease traits SSI= leaf scald severity year one, SSII= leaf scald severity year II, SSC= leaf scald severity combined over years, SCAD= leaf scald area under disease progress curve, PMSI= powdery mildew severity year one, PMSII= powdery mildew severity year two, PMSC= powdery mildew severity combined over the test years, PMAD= powdery mildew area under disease progress curve. Above diagonal correlation coefficient values of the disease traits in both diseases. Below diagonal correlation plots between any two disease traits under study.

## DISCUSSION

Developing varieties that can cope with virulent pathogens is considered as one of the most effective strategies for managing the damage caused by fungal diseases (Xu et al., 2022). Virulence of a pathogen frequently follows the gene-for-gene concept that underlines for every resistance gene in the host there is a corresponding gene conferring virulence in the pathogen (Gønneød et al., 2002; Gururani et al., 2012). Therefore, the development of resistant genotypes is a continuous process based on the identification of new sources of resistance.

In order for the spore to germinate, subsequent infection and disease development both leaf scald (Tekauz, 1991) and powdery mildew (Glawe, 2008; Scott and Punja, 2021) need enough moisture. Leaf scald specifically grows rapidly under cool and wet growing conditions. In our study, although there was a difference in disease prevalence, there was sufficient disease pressure for both diseases in the test years. The variance analysis based on BLUP mean combined over the test years for leaf scald was found to be significant for studied disease traits justifying the variation among the lines in response to leaf scald (Meles et al., 2004). In agreement with our result, significant variation for disease severity tested for two years and AUDPC among double haploid lines for leaf scald and net blotch in barley was reported (Cherif et al., 2007). The significant treatment-by-year interaction effect suggested the variability of environment and isolates that affect the response of the lines through different isolate line interactions (Yosef et al., 2017). Comparing the occurrence of the leaf scald over the test years it was more prevalent in the first year. As the date of planting in the first year was 10 days earlier than it was in the second year, earlier planting may also be considered as a probable factor explaining the higher prevalence of scald in the first year. Considering the polycyclic nature of the causal pathogen of leaf scald, an earlier planting date may have provided a temporal room for the pathogen to infect the host repeatedly during the growing season. Similar results were also described in barley (Xi et al., 2008; Zerihun et al., 2019). Hence, apart from the utilization of resistant varieties, avoiding early planting can be an alternative management strategy to reduce the damage caused by leaf scald. In addition, as leaf scald spores are dispersed by rain splash taller plants had a comparatively lower chance of being affected by the disease. Lines (Accession# 243209-A, 16866, 17148, 24639-A, 242093-A, HB-42 and EH-1847) that combine both lower scald severity and those that had slowly scalding response are capable of withstanding the reduction in yield caused by the leaf scald. Except for seven lines with moderate resistance, the rest of the lines in our experiment were in the susceptibility category for powdery mildew. However, among these moderately resistant lines for powdery mildew, two were also moderately resistant for leaf scald (HB-42 and Accn#243209) for both disease traits suggesting their

potential for multiple disease resistance. Lines of this kind are valuable sources of resistance in crop improvement activities. These lines can be used as a parental line in breeding endeavors of crossing activities to develop genotypes with multiple disease resistance. Quantitative trait loci for multiple disease resistance in wild barley on leaf scald, powdery mildew and net blotch were reported by (Yun et al., 2005). In general, disease development was maximum at the last scoring time for both diseases as a result the final disease scoring can be considered ideal in discriminating lines for the mentioned diseases especially while handling a larger set of lines.

The association between the disease traits and test years for each of the diseases was found to be highly significant and positive. In this case, the correlation coefficient between scald severity in the first and second year was ( $r= 0.82$ ). This correlation suggests resistance in the first year was resistant in the second year too despite the variation among the test years in the disease prevalence. Lines that showed low scald severity showed lower percent days for AUDPC and it is in agreement with (Paraschivu et al., 2013). Similarly genotypes with the least mildew severity exhibited the lowest AUDPC. A positive and significant correlation between severity and AUDPC disease traits for powdery mildew was in agreement with the findings of (Liatukas and Leistrumaite, 2007).

### **CONCLUSION AND RECOMMENDATION**

In the current research, the barley genotypes were studied for their performance for two major barley diseases leaf scald and powdery mildew. The finding uncovered that adjusting planting time, particularly avoiding early planting can be considered as one of the mechanisms to minimize grain yield reduction that perhaps occurs as a result of repeated infection of leaf scald thereby reducing the cycle of disease infection within the season. The varieties identified to exhibit multiple disease resistance characteristics (HB-42 and Accn# 243209) are potential breeding materials as parental lines in future breeding activities to develop varieties that combine high-yielding characteristics under multiple disease resistance backgrounds. It will

also be important to assess the response of the barley genotypes studied for the other major diseases of barley (net blotch) over different locations and years.

### ACKNOWLEDGEMENTS

The authors are thankful to the Ethiopian Biodiversity Institute and the Ethiopian Institute of Agricultural Research for the provision of barley planting materials.

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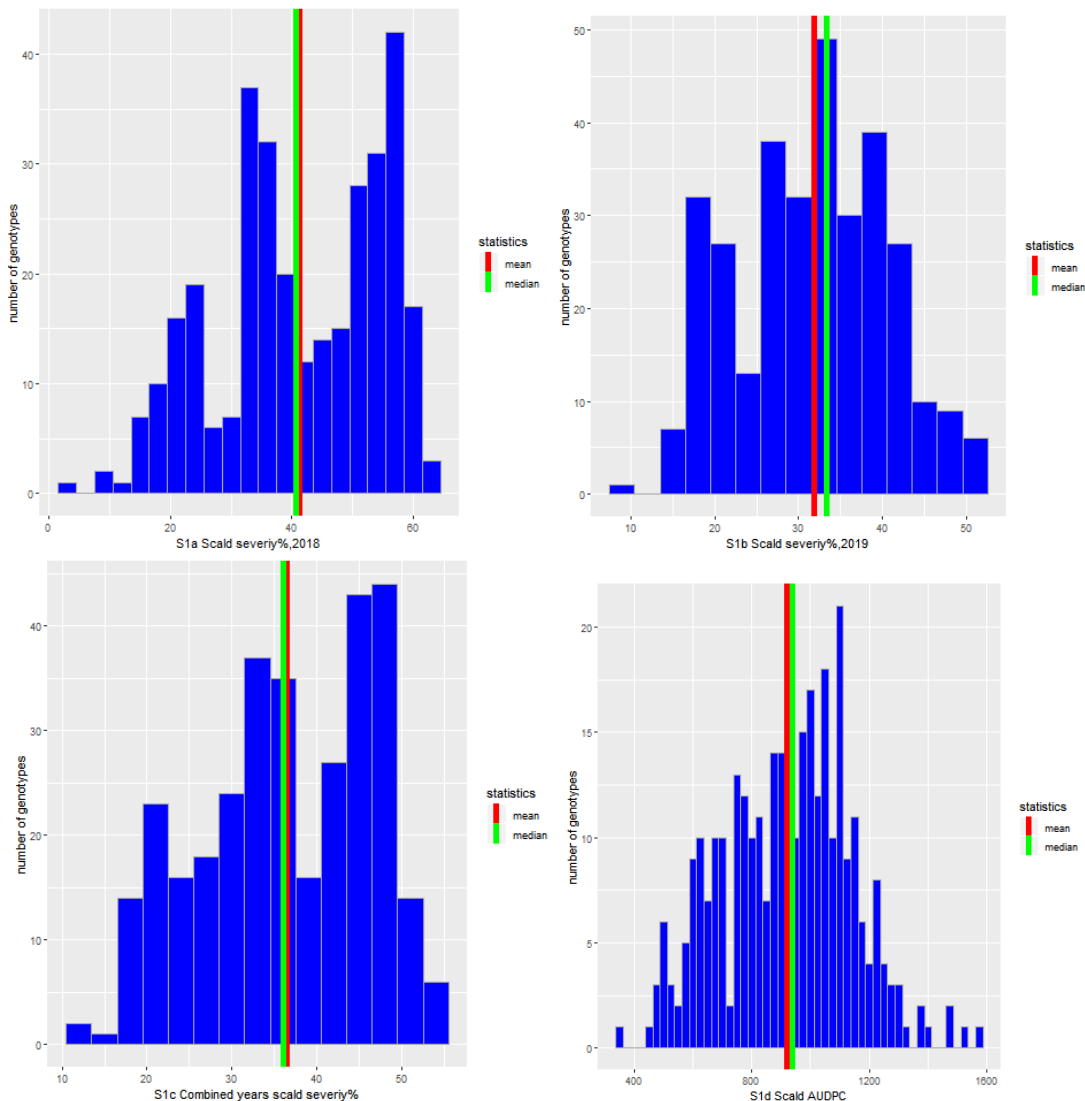
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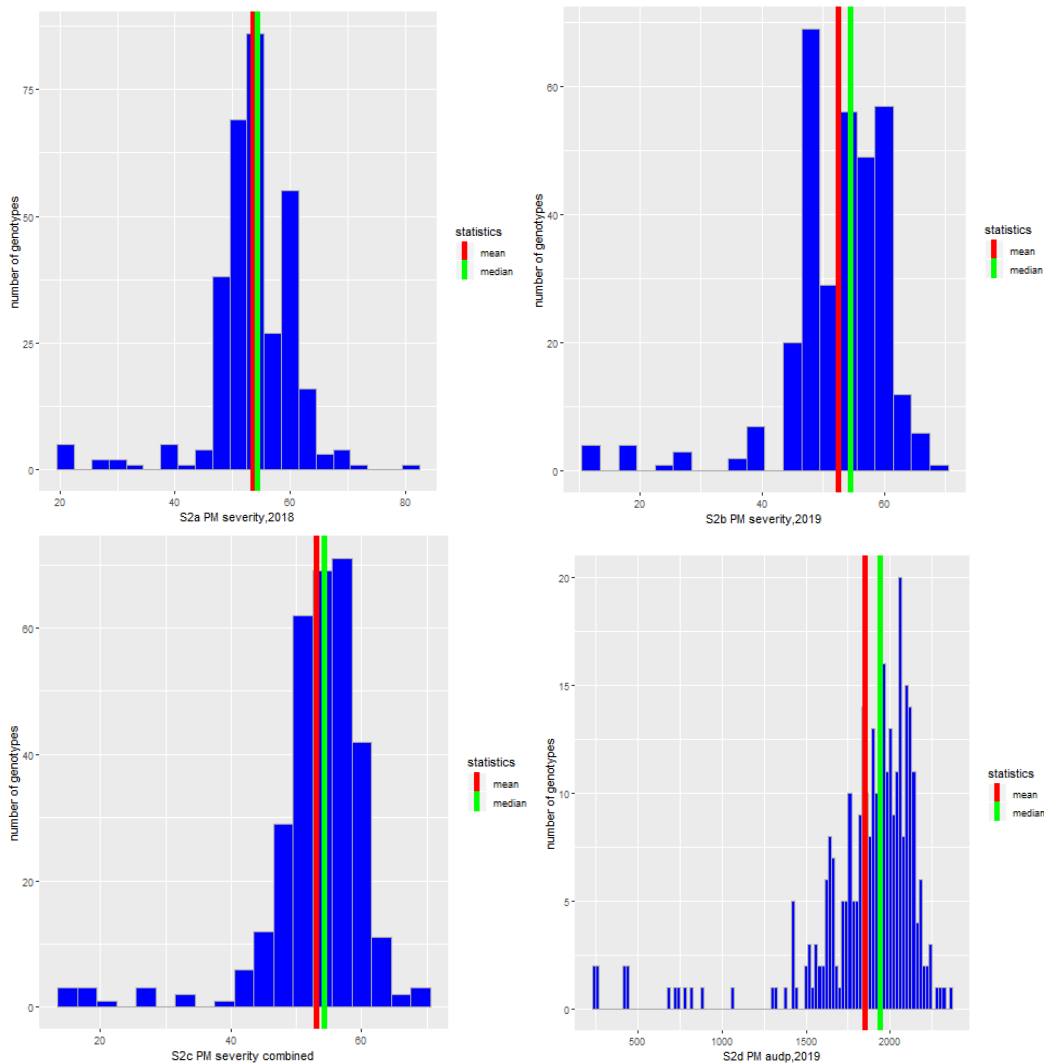
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Appendix 1. The distribution of the mean performance 320 barley lines for Leaf scald severity and AUDPC traits. S1a=scald severity % for the first year (2018), S1b=Scald severity % for the second year, S1c scald severity % combined over the test years and S1d=scald AUDPC for 2019. The x-axis disease severity percentage and AUDPC (percent days) and the y-axis number of genotypes.



Appendix 2. The distribution of the mean performance 320 barley lines for Leaf scald severity and AUDPC traits. S2a=powdery mildew severity % for the first year (2018), S2b=powdery mildew severity % for the second year (2019), S2c=powdery mildew severity % combined over the test years and S1d=powdery mildew AUDPC for 2019. The x-axis disease severity percentage and AUDPC (percent days) and the y-axis number of genotypes.

## ASSESSMENT OF THE POTENTIAL OF SEED TRAIT-BASED MODELS FOR PREDICTION OF DESICCATION SENSITIVITY OF FOREST TREE SEEDS IN ETHIOPIA

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**ABSTRACT:** The first challenge for the conservation of seeds of forest tree species is to determine their response to desiccation or seed storage behavior. This is particularly important in the tropical countries where the great portion of the forest tree species has recalcitrant seeds. The conventional experimental procedure of seed storage identification method is time consuming, requiring a germinative response and uses large amount of seeds. Estimation of seed desiccation sensitivity using seed trait-based models, thousand seed weight-moisture content (TSW-MC) criteria, and the models based on seed-coat ratio (SCR) and seed dry mass (SM) might be an alternative for the conventional experimental procedure. In this study, we assessed the potential of three seed trait-based models (i.e. TSW-MC criteria, and two other probabilistic models developed by Daws and Pelissari for prediction of desiccation sensitivity of 40 woody species with known storage behaviour from Ethiopia. The result of this study showed that the TSW-MC protocol, Daws' model and Pelissari's model to predict successfully the seed response to desiccation for 31 (77.5%), 36 (90%) and 38 (95%) of the 40 studied tree species, respectively. Once we observed a 95% efficiency rate, we have concluded that for forest tree species with unknown seed storage behavior in Ethiopia, Pelissari's model may provide more important information in a decision-making framework for the application of ex-situ seed conservation strategies.

**Key words:** Desiccation sensitivity, predictive model, seed trait, seed storage

### INTRODUCTION

Ethiopia is one of the world's biodiversity rich countries and has a very diverse set of ecosystems ranging from humid forest and extensive wetlands to the desert of the Afar depression (Gebretsdik, 2016). The flora of Ethiopia is particularly very diverse with an estimated number more than 6,000 species of higher plants, of which about 10 per cent are endemic (Hedberg et al., 2009). This biodiversity resource in general, and vegetation resources, in particular, provide many ecosystem services to the local human communities (Brandon, 2014).

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The vegetation resource of the country, however, is rapidly diminishing due to mainly deforestation and loss of habitat. Deforestation rate in the Ethiopia is estimated at about 92,000 ha per year (FAO, 2015). High emphasis is thus needs to be given to the conservation of these valuable resources to preserve their ecological benefits for the future. Conservation through genebank and/or seed banks, along with massive tree planting as restoration and plantation are some of the conservation activities that can be used to conserve the threatened species and ecosystems (Maunder et al., 2004).

The first challenge for the conservation of seeds of plant species is to determine their response to desiccation or seed storage behavior. Determination of seed storage behavior is important as, it helps to identify the type of storage conditions that are required to maintain seed viability, and to choose appropriate conservation strategy of plant genetic resources. Prior knowledge of seed storage behaviour of tree species is particularly important in the tropics where about 47% of the forest tree species have recalcitrant seeds (Tweddle et al. 2003). Long term seed storage of tropical tree species without determining the seed storage behaviour is particularly risky because there is a high probability that the seeds might be desiccation sensitive and, thus, would die when dried for storage.

So far, some protocols have been developed to classify seeds regarding their desiccation sensitivity. The familiar protocol was that developed by Hong and Ellis (1996), in which seeds are grouped as orthodox, recalcitrant, or intermediate. Although this protocol is reliable, this approach is time consuming, requiring a germinative response and uses a large amount of seeds. As a result, it is highly unlikely that all tree species will ever be identified through this procedure (Pelissari et al., 2017). An alternative approach to this common procedure is therefore needed for investigation of desiccation tolerance of targeted species. The results of previous studies have shown the potential correlates of seed desiccation sensitivity with seed traits, this includes seed mass (Pritchard et al., 2004b), seed shape (Hong and Ellis, 1997), seed moisture content at shedding (Hong and Ellis, 1998), seed allocation to physical defence and both gross and local scale habitat variables (Daws et al., 2006). Besides, different probabilistic models have also been proposed based on seed

traits to predict storage classification of forest tree seeds (For example, Pelissari et al., 2017; Wyse and Dickie 2017). Due to numerous advantages of these models compared with the conventional procedures, some germplasm banks, like Xishuangbanna Germplasm Bank of China, have been using this approach during the past two decades as a decision-making tool in the handling of species with unknown seed desiccation sensitivity Lan et al. (2014). Seed traits (e.g. seed mass and desiccation sensitivity) are, however, usually habitat-associated (Li and Pritchard, 2009; Walters et al., 2013). Evaluation of the efficiency of the seed trait-based models on typical vegetation is required before a broad usage of the models can be adopted to guide seed banking.

In Ethiopia, there exist some published studies which identifies seeds storage behaviour of forest tree species (Mewuded et al., 2017; Dagnachew et al., 2023) and there is an ongoing effort of studying seed storage behaviour of those tree species with unknown storage information by Ethiopian Biodiversity Institute. Due to the large number of tree species which requires urgent conservation action, it is highly unlikely that these efforts can generate the required information on time. This study was, therefore, initiated to assess the potential of seed trait-based models in predicting desiccation sensitivity of forest tree seeds in Ethiopia with the aim of identifying an alternative and high-throughput methods among the proposed seed trait-based models to the conventional procedure.

## **MATERIALS AND METHODS**

### **Plant material**

Matured fruit/seeds of 40 tree species from the Amhara, Benishangul-Gumuz, Gambela, Oromia and Southern Nations, Nationalities and Peoples (SNNP) regions of Ethiopia collected in years 2019-2021 were used in the study (Table 1). The altitudinal range of the specific areas from which the collection was made ranges between 448 to 2417 m.a.s.l. A change in color and fruit dehiscence was considered as an indicator of maturity during the collection.

**Table 1.** The list of studied species and the geographical information their collection sites

Species	Seed collection		Latitude	Longitude	Altitude (m.a.s.l.)
	Region	Zone			
<i>Acacia abyssinica</i> Hochst. ex Benth.	Addis Ababa	Yeka	9°02'06"	38°46'50"	2417
<i>Acacia albida</i> Del	SNNP	Hawassa	7°03'19"	38°28'06"	1691
<i>Adansonia digitata</i> L.	Benishangul	Sherkole	1036'08"	34°46'11"	770
<i>Albizia gummifera</i> J. F. Gmel.	SNNP	Hadya	7°07'38"	37°57'04"	1958
<i>Aningeria adolfi-friendericii</i> Engl. Robyns & Gilbert	Oromia	Bedele	7°45'16"	36°14'41"	2095
<i>Balanites aegyptiaca</i> (L.) Delile	Gambela	Agywa	8°16'17"	34°33'18"	451
<i>Bersama abyssinica</i> Fres.	Amhara	East Gojam	10°21'13'	37°41'34"	2351
<i>Brucea antidysenterica</i> J.F. Mill	Amhara	East Gojam	10°21'04"	37°41'41"	2408
<i>Capparis tomentosa</i> Lam.	Oromia	Jimma	7°42'37"	37°00'14"	1767
<i>Cordia africana</i> Lam.	Oromia	Jimma	7°42'37"	37°00'14"	1767
<i>Cordia simensis</i> C. gharaf, C. rothii	Oromia	Borena	4°54'52"	38°11'56"	1568
<i>Croton macrostachyus</i> Hochst. ex Delile	SNNP	Hadya	7°07'38"	37°57'04"	1958
<i>Ekebergia capensis</i> Sparrm.	Oromia	Bedele	8°20'43"	36°04'51"	1877
<i>Erythrina abyssinica</i> Lam. ex DC	Oromia	Jimma	7°39'00"	36°27'41"	1740
<i>Kigelia africana</i> (Lam.) Benth.	Gambela	Agywa	8°16'17"	34°33'18"	451
<i>Milletia ferruginea</i> (Hochst.) Bak,	Amhara	Bahir Dar	11°41'39"	37°19'04"	1780
<i>Mimusops kummel</i> A.DC.	Oromia	West Arsi	7°21'40'	38°40'10"	2097
<i>Moringa olifera</i> L.	SNNP	Goffa	6°17'59"	36°52'35"	1350
<i>Oncoba spinosa</i> Forssk.	Oromia	Bale	6°24'47"	39°46'08"	1380
<i>Pappea capensis</i> Eckl. & Zeyh.	Benishangul	Metekel	10°33'42"	36°04'31"	1792
<i>Pavetta abyssinica</i> Fres.	Benishangul	Metekel	10°32'15"	36°05'07"	1698
<i>Piliostigma thonningii</i> (Schumach.) Milne-Redhead.	SNNP	Wolita	9°55'24"	34°39'46"	1461
<i>Podocarpus falcatus</i> (Thunb.) Mirb.	SNNP	Sidama	7°06'00"	38°37'41"	1816
<i>Prunus africana</i> (Hook. f.) Kalkman.	Addis Ababa	Yeka	9°02'06"	38°46'50"	2417
<i>Prunus persica</i> (L.) Batsch	Amhara	Central	12°36'15"	37°27'59"	2186
<i>Pterocarpus lucens</i> Lepr. ex Guill. & Perr.	Benishangul	Assosa	9°55'24"	34°39'46"	1461
<i>Ricinus communis</i> Linn.	Gambela	Agywa	8°13'48"	34°16'19"	448
<i>Securidaca longipedunculata</i> Fres.	Benishangul	Metekel	6°25'05"	39°48'36"	1351
<i>Sterculia africana</i> (Lour.) Fiori	Gambela	Agywa	8°06'19"	34°44'45"	457
<i>Stereospermum kunthianum</i> Cham.	Benishangul	Assosa	9°55'24"	34°39'46"	1461
<i>Strychnos inocua</i> Del.	Benishangul	Assosa	10°36'08"	34°46'11"	770
<i>Syzygium guineense</i> (Wild) DC	Amhara	Centra	12°37'41"	37°28'54"	2378
<i>Tamarindus indica</i> L.	Benishangul	Assosa	9°55'20"	34°39'24"	1432
<i>Terminalia brownie</i> Fres.	SNNP	Goffa	6°17'59"	36°52'35"	1350
<i>Terminalia laxiflora</i> Engl. & Diels	Gambela	Agywa	8°08'50"	34°09'45"	450
<i>Trichilia dregeana</i> Sond.	Oromia	Bedele	8°20'43"	36°04'51"	1877
<i>Vangueria madagascariensis</i> J. F. Gmel.	Benishangul	Metekel	10°38'22"	36°07'30"	1450
<i>Warburgia ugandensis</i> Sprague	Oromia	Bale	6°25'05"	39°48'36"	1351



After collection, fruits/seeds were packed in cotton bags and taken to the forest seed lab of the Ethiopian Biodiversity Institute, Addis Ababa. For each species seed cleaning was done manually. Seeds were visually checked and all infested (by fungi or insects) seeds were discarded. Fleshy fruits were air-dried at room temperatures (20°-24°C) for 1 day, and cleaned within 2 days of collection by removing the fleshy pulp.

### **TSW–MC characterization**

For each species, seed moisture content and 1000 seed weight (TSW) was determined by drying about 25 cleaned seeds (103 ± 2 °C for 17 ± 1 h) following the method recommended by Rao et al., (2006) and ISTA (2019), respectively, as follows:

$$\text{Moisture content (MC) (\%)} = \left( \frac{W_2 - W_3}{W_1 - W_2} \right) \times 100$$

Where, W1 = weight of container; W2 = weight of container and seed sample before drying; and W3 = weight of container with seed sample after drying.

$$\text{Weight of 1000 seeds (TSW)} = \frac{\text{Sample weight}}{\text{Number of seeds counting}} \times 1000$$

### **Determination of water content of seed structures and ratio between tegument and dry mass of seed**

For each species, five replicates of eight seeds were dissected in order to separate the seeds into their component parts: endocarp, testa, and embryo + endosperm following Grubb and Burslem, (1998). These component parts were subsequently dried at 103°C for 17 h (ISTA, 1999) followed by mass determinations. Seed coat ratio (SCR), which is the ratio of the mass of covering structures (endocarp and testa) to the mass of the total dispersal unit was then determined for each species by using the method as described by Grubb and Burslem (1998).

### **Statistical analyses**

#### **The TSW–MC criteria for identification**

According to the TSW–MC criteria, those species having seeds with a TSW of > 500 g and MC of >30% are desiccation-sensitive (Hong and Ellis 1996). In this study, seed desiccation responses categories were then assigned for each species as follows: we considered the seed lot as “Desiccation sensitive” (DS) when

seeds had an initial moisture content (mc) >30% and a TSW >500 g. In contrast, when the initial mc <30% and TSW <500 g, we considered them as 'Desiccation tolerant' (DT). When seeds had only one of the two traits (when mc >30%, but TSW <500 g, and TSW >500 g, but mc <30%), we considered the seed as DS according to Lan et al. (2014).

### **The SCR-SM models for identification**

#### **Daws et al. (2006) model**

The likelihood of desiccation sensitivity (P(D-S)) for seeds of each of the studied 40 tree species was estimated using the equation developed by Daws et al., (2006) as follows:

$$p(D-S) = \frac{e^{3.269-9.974a+2.15b}}{1+e^{3.269-9.974a+2.15b}}$$

Where: (P(D-S)) is the likelihood of desiccation sensitivity, a is SCR and b is log<sub>10</sub> (seed mass) in gram.

To use this model, seed mass should range between 0.01 mg and 24 g, and SCR between 0 and 1. By using this model, seeds were categorized as desiccation-sensitive when P(D-S) > 0.5. Otherwise, seeds were considered as desiccation-tolerant (i.e. when P(D-S) < 0.5).

#### **The Pelissari et al. (2017) model**

The third model tested was the probabilistic model which was proposed by Pelissari, et al. (2017) as follows:

$$P = \frac{1}{1+EXP(-0.1627245*A+1.372784*B-0.4599876*C+4.348336)}$$

Where A is the water content of embryo + endosperm; B is the SCR and C is the dry weight of the seed.

According to this model, seed is classified as desiccation-sensitive if the value of p is higher than 0.5 or desiccation-tolerant if p is lower than 0.5.

Seed classification based on these three models was then compared with the information found in literature, and the potential of the three models was then compared by observing the number of times that the model predicts the seed storage behaviour accurately.

## RESULTS

Thousand seed weight, fresh seed MC, the probability of seeds of being desiccation-sensitive as determined using the Daws' et al (2006) and Pelissari's et al. (2017) models based on SCR and seed dry mass are shown in Table 2.

### **The TSW–MC criteria**

The TSW–MC criteria allowed identifying 30 of the studied species as desiccation tolerant (DT:  $MC < 30\%$  and a  $TSW < 500$  g), and 10 species as desiccation sensitive (i.e.  $MC > 30$  and  $TSW > 500$  g;  $mc > 30\%$ , but  $TSW < 500$  g; and  $TSW > 500$  g, but  $MC < 30\%$ ) (Table 1). Generally, this criterion was found to predict accurately desiccation sensitivity of 31 (77.5%) of the studied species correctly.

### **Models' predictions**

Daw's et al. (2006) model was found to predict 30 of the studied species as desiccation tolerant ( $P(D-S) < 0.5$ ), and 10 as desiccation sensitive ( $P(D-S) > 0.5$ ). This model and the TSW-MC criteria generated consistent results for 31 species (Table 2). The Daw's et al (2006) model predicted four species to be desiccation-sensitive with a  $P(D-S)$  value of  $> 0.5$  while three of these four species were predicted to be desiccation tolerant using the MC-TSW model as seed of these three tree species were small ( $TSW < 500$ g) and had low MC ( $< 30\%$ ). From the total of 40 studied species, Daw's model generally was found to predict their response to desiccation correctly for 36 species (success rate of 90%). On the other hand, Pelissari's et al., 2017 model was found to predict 28 of the studied species as desiccation tolerant ( $P(TD) < 0.5$ ), and 10 as desiccation sensitive ( $P(TD) > 0.5$ ). This model and the Daw's et al (2006) model consistently generated similar results for 37 of the 40 studied species, while Pelissari's et al. (2017) model generated similar results with the TSW-MC criteria for 31 species. From the 40 species studied, the Pelissari's et al., (2017) model correctly predicted desiccation sensitivity for 38 species (95%) (Table 2 and 3).

**Table 2.** Seed traits of the 40 tree species from Ethiopia with known seed storage behaviour that are used for evaluation of seed trait-based models.

Species	Family	MC (%)	TSW (gm)	MC-TSW model	Daws et al (2006)		Pelissari et al., 2017		DT/DS?
					Model		model		
					Probability to be recalcitrant	Possible storage behaviour	Probability to be recalcitrant	Possible storage behaviour	
<i>Acacia abyssinica</i> Hochst. ex Benth.	Fabaceae	13.3	102	DT	0.013	DT	0.222	DT	DT <sup>1</sup>
<i>Acacia albida</i> Del	Fabaceae	14.02	122	DT	0.262	DT	0.414	DT	DT <sup>1</sup>
<i>Adansonia digitata</i> L.	malvaceae	16.5	114	DT	0.019	DT	0.001	DT	DT <sup>1</sup>
<i>Albizia gummifera</i> J.F. Gmel.	Fabaceae	11.9	133	DT	0.154	DT	0.001	DT	DT <sup>1</sup>
<i>Aningeria adolfi-friendericii</i> Engl. Robyns & Gilbert	Sapotaceae	34.2	1326	DS	0.803	DS	0.798	DS	DS <sup>2</sup>
<i>Balanites aegyptiaca</i> (L.) Delile	Balanitaceae	28.6	1404	<b>DS</b>	0.015	DT	0.002	DT	DT <sup>3</sup>
<i>Bersama abyssinica</i> Fres.	Francoaceae	26.8	495	<b>DT</b>	0.656	DS	0.542	DS	DS <sup>2</sup>
<i>Bucea antidysenterica</i> J. F. Mill	<b>Simaroubaceae</b>	22.11	155	DT	0.480	DT	0.566	<b>DS</b>	DT <sup>2</sup>
<i>Capparis tomentosa</i> Lam.	Capparidaceae	27.02	201	<b>DT</b>	0.014	<b>DT</b>	0.511	DS	DS <sup>7</sup>
<i>Cordia africana</i> Lam.	<b>Boraginaceae</b>	13.3	298	DT	0.038	DT	0.006	DT	DT <sup>2</sup>
<i>Cordia simensis</i> C. gharaf, C. rothii	<b>Boraginaceae</b>	12.45	401	DT	0.281	DT	0.152	DT	DT <sup>8</sup>
<i>Croton macrostachyus</i> Hochst. ex Delile	Euphorbiaceae	11.89	65	DT	0.017	DT	0.004	DT	DT <sup>8</sup>
<i>Ekebergia capensis</i> Sparrm.	Meliaceae	35.6	152	DS	0.833	DS	0.802	DS	DS <sup>8</sup>
<i>Erythrina abyssinica</i> Lam. ex. DC	Fabaceae	11.8	225	DT	0.029	DT	0.121	DT	DT <sup>1</sup>
<i>Kigelia africana</i> (Lam.) Benth.	<b>Bignoniaceae</b>	15.06	112	DT	0.001	DT	0.002	DT	DT <sup>2</sup>
<i>Millettia ferruginea</i> (Hochst.) Bak.	Fabaceae	16.08	322	<b>DT</b>	0.602	DS	0.753	DS	DS <sup>7</sup>
<i>Mimusops kummel</i> A.DC.	Sapotaceae	22.01	225	DT	0.018	DT	0.002	DT	DT <sup>5</sup>
<i>Moringa olifera</i> L.	Moringaceae	14.78	490	DT	0.738	<b>DS</b>	0.498	DT	DT <sup>8</sup>
<i>Oncoba spinosa</i> Forssk.	<b>Flacourtiaceae</b>	13.2	58	DT	0.225	DT	0.016	DT	DT <sup>8</sup>
<i>Pappea capensis</i> Eckl. & Zeyh.	Sapindaceae	16.3	154	DT	0.451	DT	0.202	DT	DT <sup>8</sup>
<i>Pavetta abyssinica</i> Fres.	Rubiaceae	12.08	420	DT	0.003	DT	0.001	DT	DT <sup>8</sup>

Table 2. (continued)

Species	Family	MC (%)	TSW (gm)	MC-TSW model	Daws et al (2006) Model		Pelissari et al., 2017 model		DT/DS?
					Probability to be recalcitrant	Possible storage behaviour	Probability to be recalcitrant	Possible storage behaviour	
<i>Piliostigma thonningii</i> (Schumach.) Milne-Redhead.	Fabaceae	14.5	126	DT	0.025	DT	0.008	DT	DT <sup>8</sup>
<i>Podocarpus falcatus</i> (Thunb.) Mirb.	Podocarpaceae	28.1	395	DT	0.578	<b>DS</b>	0.859	<b>DS</b>	DT <sup>1</sup>
<i>Prunus africana</i> (Hook. f.) Kalkman.	Rosaceae	37.1	326	<b>DT</b>	0.487	DT	0.491	DT	DS <sup>1,10</sup>
<i>Prunus persica</i> (L.) Batsch	Rosaceae	28.07	664	<b>DS</b>	0.395	DT	0.452	DT	DT <sup>8</sup>
<i>Pterocarpus lucens</i> Lepr. ex Guill. & Perr.	Fabaceae	18.7	152	DT	0.004	DT	0.001	DT	DT <sup>8</sup>
<i>Ricinus communis</i> Linn.	Euphorbiaceae	27.9	321	DT	0.368	DT	0.056	DT	DT <sup>8</sup>
<i>Securidaca longipedunculata</i> Fresen.	Polygalaceae	10.3	305	DT	0.015	DT	0.235	DT	DT <sup>8</sup>
<i>Sterculia africana</i> (Lour.) Fiori	Flacourtiaceae	12	74	<b>DT</b>	0.016	<b>DT</b>	0.521	DS	DS <sup>2</sup>
<i>Stereospermum kunthianum</i> Cham.	Bignoniaceae	19.22	25	DT	0.365	DT	0.458	DT	DT <sup>1</sup>
<i>Strychnos inocua</i> Del.	Loganiaceae	32	458	DS	0.816	DS	0.635	DS	DS
<i>Syzygium guineense</i> (Willd.) DC.	Myrtaceae	34.2	1552	DS	0.991	DS	0.561	DS	DS <sup>9</sup>
<i>Tamarindus indica</i> L.	Fabaceae	14.5	490	DT	0.335	DT	0.197	DT	DT <sup>8</sup>
<i>Terminalia brownie</i> Fres.	Combretaceae	25.04	552	<b>DS</b>	0.002	DT	0.085	DT	DT <sup>8</sup>
<i>Terminalia laxiflora</i> Engl. & Diels	Combretaceae	22.11	415	DT	0.335	DT	0.482	DT	DT <sup>6</sup>
<i>Trichilia dregeana</i> Sond.	Meliaceae	38.09	1250	DS	0.950	DS	0.886	DS	DS <sup>4</sup>
<i>Vangueria madagascariensis</i> J. F. Gmel.	Rubiaceae	14.44	1203	<b>DS</b>	0.476	DT	0.381	DT	DT <sup>8</sup>
<i>Warburgia ugandensis</i> Sprague	<b>Canellaceae</b>	25.12	495	DT	0.407	DT	0.290	DT	DT <sup>1</sup>
<i>Ximenia Americana</i> L.	Olacaceae	36.66	1056	DS	0.995	DS	0.696	DS	DS <sup>1</sup>
<i>Zizyphus mucronata</i> Willd	<b>Rhamnaceae</b>	26.5	365	DT	0.212	DT	0.444	DT	DT <sup>1</sup>

<sup>1</sup>Girma (1999), <sup>2</sup>World agroforestry (2022), <sup>3</sup>Kamal (2014), <sup>4</sup>Anushka (2018), <sup>5</sup>Mewuded et al (2022), <sup>6</sup>Mewded et al. (2017), <sup>7</sup>Tessems (1993), <sup>8</sup> SER (2023), <sup>9</sup>Negash (2021), and <sup>10</sup>Sacandé (2004).

Cases where those models failed to predict desiccation sensitivity of the species are shown in bold font type

**Table 3.** Summary of the efficiency rate of seed trait-based models for prediction of seed desiccation sensitivity of the 40 studied species.

Model	No of spp. used	Wrong prediction	Correct prediction	Percentage
TSW-MC criteria	40	9	31	77.5%
Daws et al (2006)	40	5	35	87.5%
Pelissari et al. (2017)	40	2	38	95%

## DISCUSSION

In conservation of plant genetic resources efforts, long-term seed storage is generally considered the safest, most inexpensive and most convenient method of conservation. Most plant genetic resources are conserved by this means. However, not all seeds of woody plant species tolerate desiccation to a lower level of moisture content at which they retain their viability and can be stored in a cold room for a long period. For this reason, classification of seed storage behaviour has become a prior step in devising a suitable method of conservation for particularly those plant species with unknown seed storage behaviour.

This study tested the potential of seed trait-based models for determining seed desiccation sensitivity for 40 woody plant species from Ethiopia. For the 40 woody species with known seed storage behaviour, the TSW-MC criteria predicted successfully the seed response to desiccation for 31 (77.5%) species. The success rate of TSW-MC criteria obtained in the present study is somewhat high as compared with the accuracy level of 55% reported by Athugala et al. (2021) for selected tropical montane species in Sri Lanka, but somehow comparable with the success rate of 83% reported by Lan et al. (2014) using this criteria for tropical woody species from Southern China. As discussed by Lan et al., (2014), the TSW-MC criteria are problematic in predicting smaller or drier desiccation-sensitive seeds, and this could explain the present result since three (60%) of the five cases that this model fails to predict desiccation sensitivity in the present study had a small and dried seeds.

As a rule of thumb, desiccation sensitive seeds are larger and have higher moisture content at dispersal. Accordingly, seed mass and initial moisture content (TSW-MC criteria) was described by Hong and Ellis (1996) as a predictive indicator for the response to desiccation tolerance. Although having some limitations, this protocol was used successfully to predict seed storage behaviour of woody species from various tropical regions; Woody species from tropical montane species in Sri Lanka (Athugala et al., 2021), for Caribbean native tree species (Mattanna et al., 2020) and for woody species from Southern China (Lan et al. 2014). The SCR–SM models provided by Daws et al. (2006) and Pelissari et al. (2017) successfully predicted the seed response to desiccation for 36 (90%) and 38 (95%) of the woody species, respectively. This result is in accordance with the success rate (88%) achieved using Daws et al. 2006 model for 101 woody species from Southern China (Lan et al., 2014), and that of 92% reported by using Pelissari et al. (2017) model for 66 Brazilian tree species (Pelissari et al., 2017). The similarity of the success rates observed from the result of present study for those 40 tree species sampled from a large range of altitudinal differences, with that of previously reports suggested that the SCR–SM models are a reliable predictive method for Ethiopian woody species.

As described by Hill et al. (2012), the ratio between the dry weight of the tegument and endocarp (SCR) can be a better predictor than the seed itself. Desiccation sensitive seeds, when compared with desiccation tolerant seeds, usually have a thick seed coat (Pritchard et al. 2004b). These results corroborate those of Pritchard et al. (2004b), Daws et al. (2006) and Hamilton et al. (2013), once the mass allocation on the external seed layer becomes a desiccation tolerance indicator (Pritchard et al. 2004a). According to Daws et al. (2006), the SCR reduces the chances for large orthodox seeds, with high mass to be classified as recalcitrant, showing that SCR for orthodox seeds is high and identified as a good predictor of desiccation-tolerance.

The result of this paper showed that the protocols based on SCR is a reliable predictor for Ethiopians woody species seed classification regarding desiccation tolerance and storage as reported by Pelissari et al., (2017),

Lan et al., (2014) and Daws et al. (2006). In particular, once we observed a 95% efficiency rate for the studied species which were collected from a diverse set of environments, we have concluded that for forest tree species with unknown seed behavior in Ethiopia, the model provided by Pelissari et al., (2017) may provide more important information in a decision-making framework for the application of *ex-situ* seed conservation strategies. However, although the SCR–SM model is robust and more reliable than the TSW–MC criteria, a seed mass of 0.01 mg to 24 g is required (Pelissari et al., 2017, Daws et al. 2006), and in many species, data for SCR are not available. In which case, we recommend TSW–MC criteria as a practical tool to predict seed storage behaviour of woody species. Besides, TSW–MC criteria, with the observed 77.5% efficiency rate, may still be a very important tool for decision making in cases where large collection of forest tree seeds with unknown seed storage behavior are made, and quick decision regarding the choice of appropriate conservation strategies for each of the collected species has to be made.

### **CONCLUSIONS AND RECOMMENDATION**

In this study, the potential of three seed trait-based models in predicting seed storage behaviour of Ethiopian forest tree species was made by comparing the result obtained from each model to the report from published material that used the usual long experimental procedure. Although some additional studies with other tree species might be needed for the general acceptance of these models, the result of this study has witnessed that these models are robust and reliable for predicting seed storage behaviour of tree species with unknown seed storage behaviour in Ethiopia. These findings might particularly be important to demonstrate the potential of these models, if used in the future, as an alternative to the usual long experimental procedures in decision making regarding the choice of appropriate *ex-situ* conservation strategy for tree species with unknown seed information.



## ACKNOWLEDGEMENTS

The authors would like to acknowledge the Forest and Range Land Plants Biodiversity Directorate of the Ethiopian Biodiversity Institute for funding this research work. We also thank Miss Helen for her support during data collection in the forest seed lab.

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**POPULATION STRUCTURE AND REGENERATION OF *POUTERIA*  
*ADOLFI-FRIEDERICII* TREE SPECIES IN MOIST EVERGREEN AFROMONTANE FORESTS  
OF SOUTH WEST ETHIOPIA**

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**ABSTRACT:** Knowledge of tree population structure and regeneration status is very important to understand the reproductive and recruitment potential of selected indigenous tree species. The aim of this study was to investigate the population structure and regeneration status of *Pouteria adolfi-friedericii*. The study was conducted in four natural forests of Yayu, Bonga, Bebeke, and Masha, in south western Ethiopia. A systematic sampling method was used to collect vegetation data. Ten transects of 160 m length were laid out in each forest at 100 m interval along the slope gradient and quadrats (size: 20 m×20 m each) were laid at 50 m interval along each transect line. A total 120 quadrats were used for vegetation data collection. Sub quadrats (size: 5 m×5 m each) were established at four corners and in the center of each main quadrat to collect data on regeneration. From each main quadrat, the DBH and total height of the species were measured by using a diameter tape and clinometer, respectively. The size class distribution of the species showed irregular patterns across the forests. The regeneration status of *P. adolfi-friedericii* is “good” in Masha and Bebeke, and “fair” in Bonga and Yayu forests. The population structure of the species varied across the inventoried forest sites.

**Keywords:** Population structure, *Pouteria adolfi-friedericii*, Regeneration, Seed production

## INTRODUCTION

Native trees in tropical forests, are severely affected by a complex set of causes. Anthropogenic activities have been modifying tropical forest land cover for food and energy production (Takahashi et al., 2017). Indigenous tree species population are declining from their natural ranges, especially for non-industrial plantations, and there has been little attention devoted to the practice and domestication of such tree species (Nichols et al., 2006).

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Ethiopia has a wide variety of natural conditions that resulted in the existence of heterogeneous flora and fauna, which made the country one of the major centers for biodiversity (Woldemariam, 2003; Alemayehu et al., 2005). The southwestern part of Ethiopia is covered by moist, evergreen montane forests and has a high concentration of native tree species important in providing timber and non-timber products of the forest (Chilalo and Wiersum, 2011; Senbeta, 2014). The moist forest ecosystem is the most diverse ecosystem in composition, structure and habitat types consequently it is rich in biodiversity with a number of endemic species. Some of the characteristic plant species of the forests include; *Pouteria adolfi-friedericii*, *Albizia gummifera*, *Prunus africana*, *A. schimperina*, *Blighia unijugata*, *Cassipourea malosana*, *Celtis africana*, *Croton macrostachyus*, *Ekebergia capensis*, *Euphorbia ampliphylla*, *Ficus sur*, *Ilex mitis*, *Macaranga capensis*, *Olea capensis ssp. welwitschii*, *Polyscias fulva*, *Schefflera abyssinica*, *Sapium ellipticum*, and *Syzygium guineense ssp. Afromontanum* (Friis, 2010).

The existing knowledge on the extent of the montane moist forest ecosystem is limited though there are studies on the composition and structure some forest vegetation that exist in this ecosystem (Woldu et al., 1989; Yeshitila, 1997). The moist evergreen montane forest consists of high forests of the country mainly the south west forests. At any site, the plant diversity is influenced by species abundance and distribution patterns (Palit et al., 2012). Species wise, some studies have been conducted to investigate the population dynamics of the montane moist forest of Ethiopia (Hadera, 2000; Tesfaye et al., 2019, Tadesse et al., 2023). *Pouteria adolfi-friedericii* is among the timber tree species found in moist evergreen Afromontane forests within altitudinal range of 1350 – 2450 m.a.s.l in Ethiopia (Hedberg et al., 2003). It is usually found in areas of high-rainfall and commonly found in the Illubabor, Kafa, and Bench-Maji zones (Bekele, 2007). For this study, *P. adolfi-friedericii* was selected because of its high wood quality and economical importance. In the selected study sites, the species is highly exploited by farmers' and loggers for domestic use and trade, without any consideration of its future sustainability. Hence, due to heavy exploitation, this tree species is at risk in Ethiopia. The objectives of this research were to compare the population of *P. adolfi-friedericii*

based on their abundance, regeneration status and reproductive phenology in the selected study sites to establish seed production area and investigate the population structure and natural regeneration status of *P. adolfi-friedericii* in southwestern forests.

## MATERIALS AND METHODS

### Description of the study area

The study was conducted in Bonga, Bebeke, Masha and Yayu natural forests of south western Ethiopia. Bonga forest is located in the in Kafa zone; Masha Forest is situated Sheka zone and Bebeke forest in the Bench Maji zone of the Southern Nations, Nationalities and Peoples Regional State (SNNPRS). Yayu forest, on the other hand, is located in the Illubabor zone of the Oromia National Regional State (Figure 1).

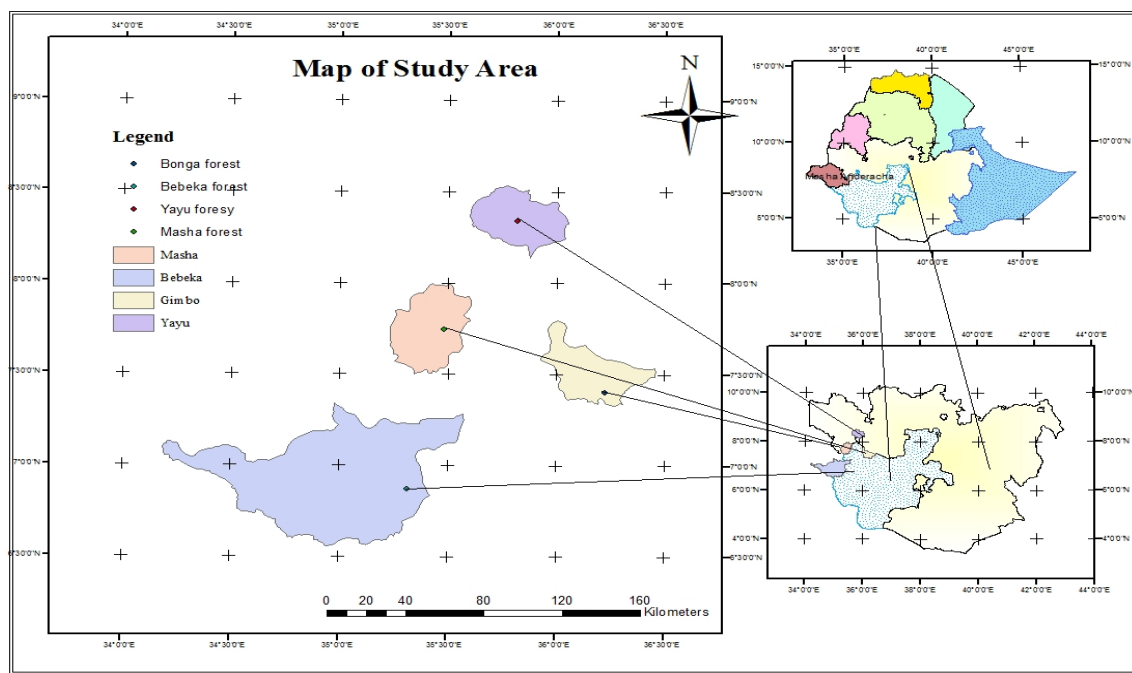
Bonga forest lies between 7°00'–7°25'N latitude and 35°55'–36°37' E, at altitudes between 1000 - 3400 m.a.s.l. The average annual temperature in Bonga is around 19°C, with the warmest month being March (average temperature of 21.3°C) and the coldest month being August (average temperature of 16.5°C). The average annual rainfall in Bonga is about 1,400 mm, with the rainy season occurring from March to September.

Masha forest is located in the geographic range of 7°24'–7°52'N latitude and 35°13'–35°35'E longitude with altitudinal range between 1700 -3000 m.a.s.l. The average annual temperature in Masha is around 14°C, with the warmest month being March (mean temperature of 15.9°C) and the coolest month being August (average temperature of 12.3°C). The average annual rainfall in Masha is about 2,400 mm, with the rainy season lasting from March to October.

The Bebeke forest is located within 07°16' N and 36°15' E longitude with an altitudinal range of 1000 - 1350 m.a.s.l. The average annual temperature in Bebeke is around 25°C, with the warmest month being March (average temperature of 26.8°C) and the coolest month being August (average temperature of 23°C).

Yayu forest is lies between 8°21'–8° 26' N latitude and 35°45'–36°3' E longitude with an altitudinal range of 1200 -2000 m. a.s.l. The average annual temperature in Yayu is around 16°C, with the warmest month

being March (average temperature of 18.1°C) and the coolest month being August (average temperature of 14.1°C). The average annual rainfall in Yayu is about 2,100 mm, with the rainy season occurring from March to October.



**Figure 1.** Map of the study area

### Soil property

The soils of the study area vary in color and type depending on the topography and types of the parent materials. Most parts of the southwestern Ethiopia is dominated by Cenozoic and Proterozoic volcanic sediments (Schlüter, 2008). In Bonga, the soil is characterized as deep red to brown red, lateritic loams or clay loams of volcanic origin with high or medium fertility (Schmitt, 2006). Whereas the commonly observed soils in Masha vary in color from black to red. Nitisols, Vertisols, Fluvisols and Cambisols are the dominant soils types in the area. The dominant soil type in Yayu Forest is Nitisols. These types of soils are deep, reddish-brown and clayey with relatively high organic matter content. Nitisols have a crumb and/or sub-angular structure and well drained. The soils of Bebeke forest are sandy loam, moderately drained and reddish soil with 15-20 cm thick litter and humus.

## Sampling Design

### Population structure

The selection of the natural forests of the study was made after considering the potential of the populations of the species studied and taking into account previous field research experience, relevant literature reviews and input from experts and community leaders in the relevant woredas and farmer associations. In addition, a reconnaissance survey was conducted to determine the representative habitats of the research area, the spacing between transect lines, and the location of plots along each line. Ten transect lines were constructed every 100 m along the slope using a systematic sampling technique to collect vegetation data.

Sample quadrats of 400 m<sup>2</sup> (20 m×20 m) each for tree and sapling were laid out at each 50 m interval in each transect line. In each main quadrat, sub quadrat of 25 m<sup>2</sup> (5 m×5 m) for seedling of the target tree species were laid out at four corners and in the center. A total of 120 (30 quadrats for each site) were sampled for *P. adolfi-friedericii* at the Bonga (Adela site), Masha (Gorashewi site), Yaya (Durani site), and Bebeke (Duduka site). The target species was not found in all surveyed areas, possibly due to altitude and other ecological conditions.

## Data Analysis

### Population structure

Both height and DBH data of the species were entered, cleaned, organized and summarized in Microsoft Excel and SPSS software. Nine DBH classes (i.e., <10cm, 10.1 - 20 cm, 20.1 - 30 cm, 30.1 - 40 cm, 40.1 - 50 cm, 50.1 - 60 cm, 60.1 - 70 cm, 70.1 - 80 cm, >80 cm) were established based on the DBH size ranges measured for the species.

Basal area (BA) was calculated using the formula:

$$\text{Basal area} = (A) = \pi r^2; \text{ where, } \pi=3.14$$

Density estimates obtained from transects were used to calculate the number of individuals of the species in the study area. It was a count of the number of individuals of the species within the quadrat on hectare



basis (Kent and Coker, 1992). Afterwards, the sum of individuals of the species was calculated and analyzed in terms of species density per hectare.

$$D \text{ (density)} = \frac{\text{number of stems of species counted}}{\text{sample area}}$$

Individual trees having height  $\geq 2$  m and DBH  $\geq 10$  cm within sampling quadrats were collected and analyzed by classifying into seven height classes (2 -10 m, 10.1 - 19 m, 19.1 - 28 m, 28.1 - 37 m, 37.1 - 46 m, 46.1 - 55 m, >55 m). Population structure was summarized using histograms of diameter size classes. ANOVA was used to test for difference in basal area, DBH, height and number of individuals per hectare among forests.

### **Regeneration**

The regeneration status of *P. adolfi-friedericii* in each forest habitat was analyzed by comparing the population density of seedling, sapling and matured trees (Dhaulkhanda et al., 2008 ; Gebrehiwot and Hundera, 2014) as follows: 1)“good” regeneration, if density of seedling > sapling > mature tree; 2)“fair” regeneration, if density of seedling > sapling < mature tree; 3)“poor” regeneration, if a species survives only in the sapling stage, but not as seedlings; 4)“none”, if a species is absent both in sapling and seedling stages, but present as mature; and 5)“new”, if a species has no mature, but only sapling and/or seedling stages. All forests were compared in terms of their regeneration status and the best forest habitat was recommended for seed production.

## **RESULTS**

### **Population Structure**

#### **DBH, Density and Basal area**

A total of 142 individuals of *P. adolfi-friedericii* were recorded in 56 plots out of 120 plots in sampled forests. Out of 30 plots in each site, in Adela site (Bonga), 27 trees were recorded in 11 plots; in Gorashewi

site (Masha), 61 trees were recorded in 17 plots; in Durani (Yayu), 22 trees were recorded in 14 plots and in Duduka site (Bebeka), 32 trees were recorded in 14 plots.

The density, DBH and height of *P. adolfi-friedericii* were significantly higher in Gorashewi site natural forest than Adela, Duduka and Durani sampled natural forests (Table 1). The highest number of individuals by diameter class was recorded in 20.1-30 cm, >80.1 cm, 30.1-40 cm and <10 cm for Bonga, Masha, Yayu and Bebeka forests, respectively (Figure 2).

**Table 1.** Density, DBH, Height, and Basal area of *P. adolfi-friedericii* among sampled natural forests.

Natural Forests	Density (trees/ha) (Mean $\pm$ SD)	DBH (cm) (Mean $\pm$ SD)	Height (m) (Mean $\pm$ SD)	Basal area (m <sup>2</sup> /ha)
Adela (n=30)	61.29 $\pm$ 0.59	29.00 $\pm$ 1.11	25.48 $\pm$ 1.49	62.53 $\pm$ 1.126
Gorashewi (n=30)	95.24 $\pm$ 0.58	64.73 $\pm$ 1.46	26.12 $\pm$ 0.91	589.00 $\pm$ 1.126
Durani (n=30)	78.51 $\pm$ 0.72	38.02 $\pm$ 0.7	24.72 $\pm$ 0.71	65.21 $\pm$ 0.548
Duduka (n=30)	57.09 $\pm$ 1.00	44.64 $\pm$ 1.70	29.9 $\pm$ 1.38	134.99 $\pm$ 1.100
<i>P</i> value	<0.05	<0.05	<0.05	<0.05

SD=standard deviation; n=number of plots

The DBH class distribution patterns of *P. adolfi-friedericii* in Duduka was characterized by higher individuals at middle stage than mature aged population, in which the density of individuals in the lower and middle DBH class is very high but becoming lower in the highest DBH classes. The height class distribution showed that large number of individuals exhibit middle size classes (between size classes 4 and 6 or from 29 m to 55 m with some individuals characterized by lower height class and very few individuals by large size classes (Figure 2). The maximum height value was recorded for Masha forest.

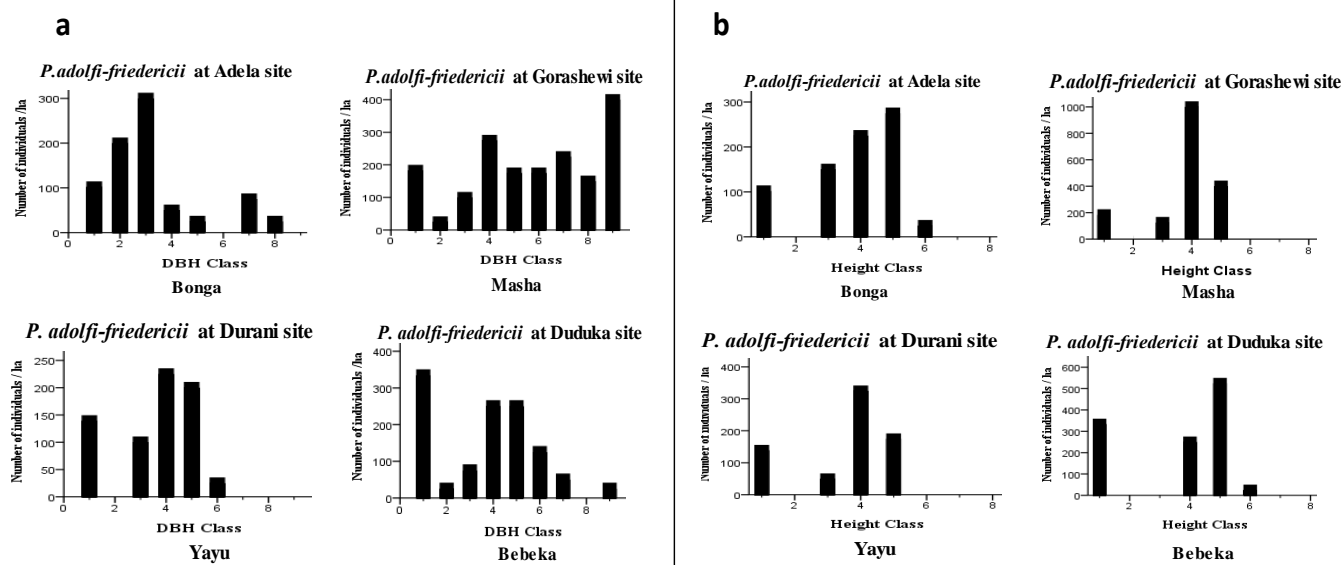


Figure 2. Population structure of *P. adolfi-friedericii* in the four studied sites

### Regeneration status

Seedling density was higher for Duduka forest while sapling and mature trees density was higher for Gorashewi forest (Figure 3). This implies that regeneration status of the species can be taken as good status in Gorashewi and Duduka forests as seedling > sapling > mature tree.

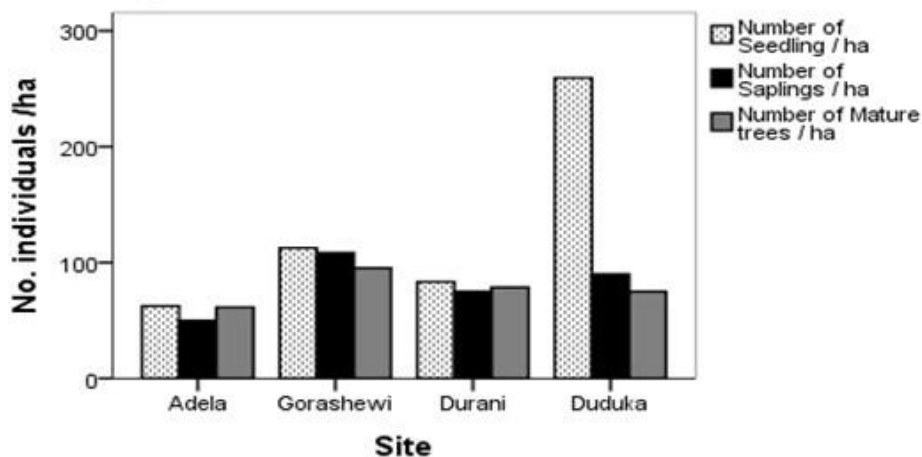


Figure 3. Regeneration status of *P. adolfi-friedericii* in the studied sites.

On the other hand, it is fair regeneration for Adela and Durani forests with seedling > sapling < mature trees. These representative figures show that the seedling, sapling and mature tree of *P. adolfi-friedericii* in

different samples natural forests. In Duduka and Gorashewi site the natural regeneration status represents good regeneration and recruitment which seedling > sapling > mature tree. While, in Adela and Durani site natural forests fair regeneration was observed.

Thus, the present study showed that population structure and regeneration status of *P. adolfi-friedericii* are good in Gorashewi forest which makes it suitable for seed production primarily. Following Gorashewi, Duduka forest is also suitable for seed production as observed from the population structure and regeneration status of the species. On the contrary, the population structure and regeneration status at Adela and Durani forests indicated insufficient number of individuals and absence in some diameter and height classes hence the regeneration status in these forests was ranked as fair.

## DISCUSSION

This study showed that *P. adolfi-friedericii* has a clustered distribution pattern in the forest. A total of 142 individuals of *P. adolfi-friedericii* were recorded in 56 plots out of 120 plots in the sampled forests. The distribution of this tree species was influenced by slope and elevation. This result is consistent with an earlier study which reported that elevation and slope influence the distribution of *P. adolfi-friedericii*, the species being more abundant at higher elevations and on north- and east-facing slopes (Asefa et al., 2017). DBH class distribution of individuals showed an irregular pattern in Adela and Gorashewi forests in which they were distributed differently in almost all classes. The reason for such irregularities could be anthropogenic impacts.

The DBH class distribution patterns of *P. adolfi-friedericii* in Duduka site natural forest was characterized by higher individuals at middle stage than mature aged population, in which the density of individuals in the lower and middle DBH class is very high but becoming lower in the highest DBH classes even nothing in some DBH classes. According to Gebrehiwot and Hundera (2014) this pattern showed that there is selective cutting of the species for different purposes like for construction and fuel. Mean density, DBH, and basal area significantly varied among forests ( $P < 0.05$ ). This result is consistent with the findings of

Teshome et al. (2019) who reported that the species is distributed in clusters, with higher densities in some areas than others.

Similar to the DBH class distribution, there are missing individuals at some height classes across all forests. The absence of large individuals or their presence in few numbers in a forest might be associated with the selective cutting of species for various purposes such as construction, firewood etc. (Gebrehiwot and Hundera, 2014).

The height distribution patterns of *P. adolfi-friedericii* in Gorashewi natural forest was characterized by higher individuals at middle stage than young and mature aged population. This result is consistent with the outcome of Ngomanda et al. (2019), which examined the tree species composition and structure of a forest in Gabon, including *P. adolfi-friedericii*.

The regeneration status of the *P. adolfi-friedericii* tree species at the study sites is satisfactory, indicating good regeneration status, but the target tree species falls below reasonable regeneration status at the Adela site. Prior studies on the regeneration of *P. adolfi-friedericii* indicated that it normally exhibits low rate of regeneration. In two forest reserves in Côte d'Ivoire, Kouamé et al. (2014) investigated the regeneration status of *P. adolfi-friedericii* and discovered that the species rarely regenerates. Koffi et al. (2016) also reported that the species had a very poor rate of regeneration, with only a few seedlings being seen in their study area. The restricted recovery, according to these studies, was caused by habitat fragmentation and overexploitation. Overall, these studies suggest that *P. adolfi-friedericii* is experiencing limited regeneration in its native range, likely due to habitat fragmentation, overexploitation, and other human activities.

### CONCLUSIONS AND RECOMMENDATION

The population structure and regeneration status of *Pouteria adolfi-friedericii* in different natural forests in south-western Ethiopia showed that the occurrence of the species is relatively low in all the selected natural forests. The sites, characterized by fair regeneration of *P. adolfi-friedericii* (Adela and Durani), showed that the growth, survival and reproductive potential of the species is at risk. Therefore, urgent priority needs to

be given to conservation and management. The presence of good regeneration potential in Gorashewi and Duduka forests indicated the species' suitability for the environment. In general, due to the population structure and regeneration status of *P. adolfi-friedericii*, the Masha Forest is more suitable for the establishment of seed production areas.

### ACKNOWLEDGEMENT

We would like to extend our heartfelt thanks to the United Nations Development Program (UNDP) and the Ethiopian Environment and Forest Research Institute (EEFRI) for the financial assistances, material support and cooperation during data collection. Our appreciation and gratitude also go researchers at Jimma Environment and Forest Research Center for their effective facilitation during data collection. We would also like to acknowledge our colleagues and others who helped us during our research work. We thank Jimma University College of Agriculture and Veterinary Medicine for facilitating the entire research work.

### Declaration

The manuscript is part of an MSc thesis of the first author.

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## PRODUCTION SYSTEM OF INDIGENOUS CHICKENS IN PASTORAL AND AGRO-PASTORAL DISTRICTS OF SOUTH OMO ZONE, SOUTH ETHIOPIA

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**ABSTRACT:** In Ethiopia, poultry production offers considerable opportunities in terms of generating employment opportunities, improving family nutrition, empowering women and ultimately ensuring household food security. The objective of this study was to characterize the production system of indigenous chickens in pastoral and agro-pastoral districts of South Omo Zone. A total of three districts and seven kebeles were purposively selected based on chicken population number and production potential of the selected areas. A total of 81 households were randomly selected for characterization of the production system. Data was gathered using semi-structured questionnaire, and field observations. The study showed that most of the household heads were male (70.3%). The average flock size of local chicken was  $13.3 \pm 0.4$  per household and the flock structure includes pullets (30.8%), layers (24.1%), cocks (17.5%), chick (16.6%) and cockerels (11.0%). Traditional chicken management system was the dominant production system (82.7%) practiced in the areas. The major feed sources for indigenous chickens were open scavenging and seasonal feed supplementation. Maize and sorghum grains as well as household leftovers were major supplements used. Newcastle disease was the most common diseases in study districts. The chicken populations have good potential for egg and meat production and the reproductive performances was also reasonable under the existing limiting environmental factors. The major constraints in the districts were disease, predator and feed shortages. Studying the production system of indigenous chickens can be used as first step to design conservation and improvement strategies, and contribute to sustainable utilization of indigenous chickens at scavenging environment.

**Keywords:** Indigenous chicken, characterization, production system.

### INTRODUCTION

Poultry production in Ethiopia offers considerable opportunities in terms of generating employment opportunities, improving family nutrition, empowering women (especially in rural areas) and ultimately ensuring household food security (FAO, 2019). Extensive poultry production is often the domain of poor women as it requires little initial investment and does not usually conflict with other household duties (FAO,

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2019). Poultry production system in Ethiopia is characterized by small flock sizes, low input, low output, and periodic devastation of the flock by disease. There are about 41.35 million chickens; of which 78.04% are local ecotypes (CSA, 2022). This indicates the relevance of indigenous chicken as principal potential farm animal genetic resources of the country.

Indigenous chicken contributes high quality animal protein in the form of eggs and meat for home consumption as well as for sacrifices and are also easily managed by all even the poorest of the poor including women and children. These chicken ecotypes have been reported to adapt very well to the traditional small-scale production system of the rural community (Petrus, 2011). They are known to possess desirable characters such as thermo tolerant, resistant to some disease, good egg and meat flavor, hard eggshells and high dressing percentage (Aberra, 2000). In addition, they have fast generation interval and high reproductive rate as they are prolific, easy to rear and their output can be generally expanded more rapidly and easily than that of other livestock (Dhuguma, 2009).

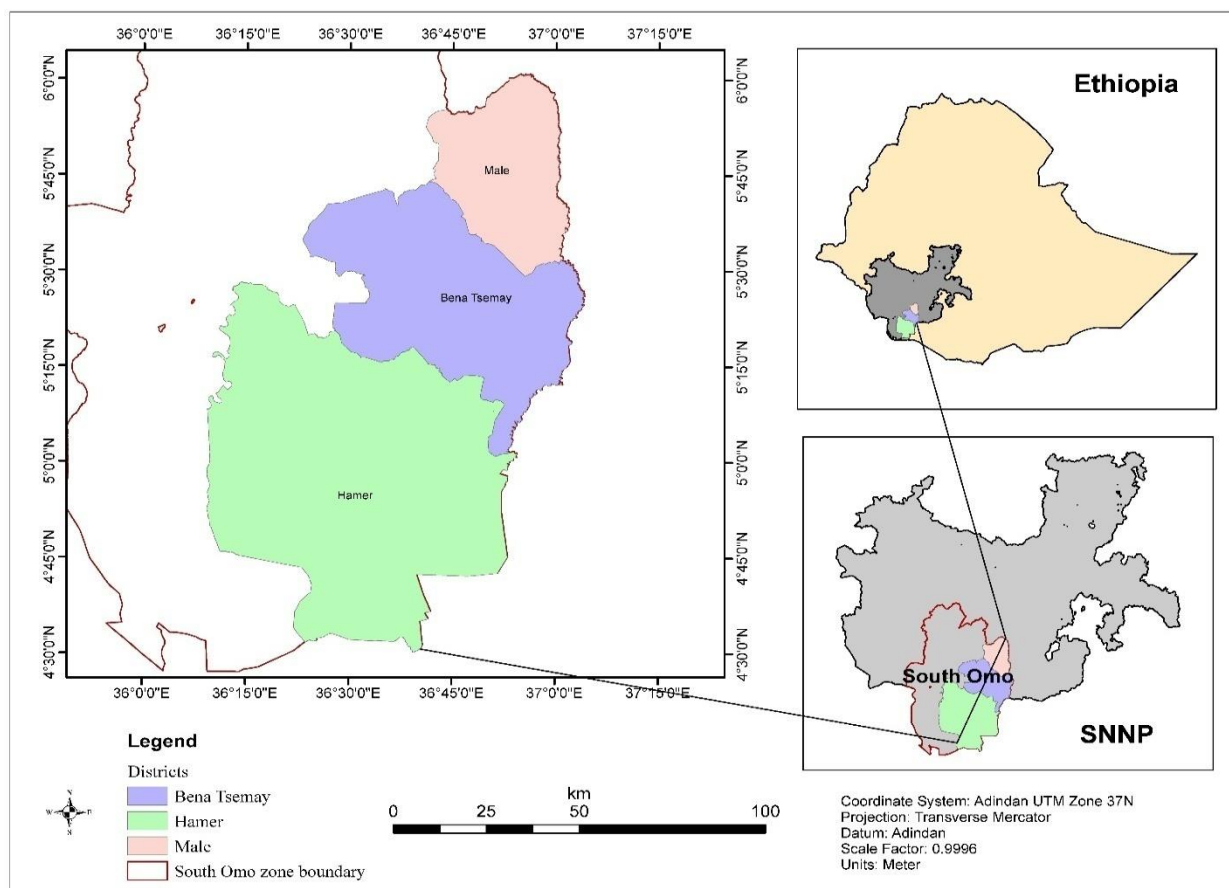
South Omo Zone is rich in indigenous chicken resources but the production system of indigenous chickens was not well studied and documented in pastoral and agro pastoral areas of the South Omo zone. The objective of this study was to characterize the production system of indigenous chickens in South Omo Zone. Characterization indigenous chicken's production system is imperative to have comprehensive data and information on socioeconomic aspects of owners, flock structure, production system, management and mobility, feeds and feeding management, productive and reproductive performance, health and production constraints.

## **MATERIALS AND METHODS**

### **Description of Study Area**

South Omo zone is located in South-West of Southern Nations, Nationalities and Peoples regional state (SNNPR). According to the South Omo zone agricultural department (2018), the zone roughly lies between 4° 43' N to 6° 46' N latitude and 35° 75' E to 37° 07' E longitude. It is bordered with Keffa zone and Konta

special district in the North, Gamo Gofa zone and Basketo special district in North East, Kenya in South, Segen Zuria People's zone in the East, Oromiya region (Borena zone) in South East, and Bench Maji zone in the West and North West (Figure 1).



**Figure 1.** Map of the study area.

The information obtained from Zone agricultural department (2018) indicated that the total area of the zone is estimated to be 22,835.80 sq.km, which shares 20.94% of the total area of SNNP region. The population size of the zone, according to the 1999 E.C population census projection result is estimated to be 790,798 accounting nearly 4% of the total population of the region. The average population density of the zone is 34.6 persons per sq.km. This zone consists of 16 ethnic groups that have their own distinct geographical location, language, culture, and social identities.

## Sampling techniques

In collaboration with the zonal livestock office, study districts were selected considering chicken populations, agroecology and potential area for poultry production. Accordingly, two agro-pastoral (Benatsemay and Male) and one pastoral (Hamer) districts were selected. Sampling sites (kebeles) were selected from each sample district based on the chicken population size data obtained from the respective districts of livestock development office. Accordingly, three kebeles from Benatsemay (Aladuba, Luka and Kako), two kebeles in Male (Boshkoro and Gudo) and two kebeles in Hamer (Erayaunbule and Senbele) districts were selected for the study. In totally 81 households (35 in Benatsemay, 30 in Male and 16 in Hamer) districts were selected based on population size of study the districts. Households with minimum number of two chickens and had prior experience in local chicken production were selected.

## Data collection

Data were collected by administering a semi-structured questionnaire, focus group discussion, and field observation. A modified questionnaire was prepared by FAO guideline (FAO, 2012). The questionnaire was used to collect information household characteristics, livestock species composition, flock structure, production system, management and mobility, feeds and feeding management, productive and reproductive performance, identification of major diseases and production constraints. Semi-structured questionnaires were also administered to randomly selected pastoralists and agro pastoralists in selected kebeles who were interviewed for the household survey.

## Data Managements and Analysis

The collected data was checked, coded and entered to SPSS (2009) software for analysis. Indices were employed to calculate the rank of the production constraints and class of chickens receiving supplementary according to the following formula:

Index =  $\Sigma$  of [3 for rank 1 + 2 for rank 2 + 1 for rank 3] divided by  $\Sigma$  of [3 for rank 1 + 2 for rank 2 + 1 for rank 3] for rank.

## RESULTS

### Socioeconomic status

Household characteristics and socio-economic aspects of the sampled households are presented in (Table 1). The majority (70.3%) of the interviewed households in the study area were male headed. The age of the majority of the respondents (95.7%) falls under 50 years old, which is the active age group to undertake chicken production effectively. The educational status of the respondents was 62.7, 13.3, 16.8 and 7.2% for illiterate, read and write, grade 1 to four and grade five to eight class attendants, respectively. The result revealed that most of the respondents participated in this study were illiterate. The average family size of the households was  $5.84 \pm 0.48$ . The results show that there are no significant differences ( $P < 0.05$ ) between the study districts of the family size.

### Livestock species composition

The average livestock species composition of the study area is presented in Table 2. Respondents in Hamer district had significantly higher number of cattle, sheep, goat and bee colony holding than respondents in Benatsemay and Male districts. However, they had significantly ( $P < 0.05$ ) lower number of chickens in Hamer district compared to Benatsemay and Male districts.

**Table 1.** Socioeconomic characteristics of the sampled households in the study areas.

Variables	Districts							
	Benatsemay (n=35)		Male (n=30)		Hamer (n=16)		Overall Total (n=81)	
	N	%	N	%	N	%	N	%
<b>Sex structure</b>								
Male	25	71.4	25	83.3	9	56.2	59	70.3
Female	10	28.6	5	16.7	7	43.8	22	29.7
<b>Age structure</b>								
15-30	16	45.7	11	36.7	3	18.8	30	33.7
31-40	13	37.1	9	30	8	50.0	30	39.0
41-50	5	14.3	7	23.3	5	31.2	17	22.9
51-60	1	2.9	1	3.3	-	-	2	2.1
61-70			2	6.7			2	2.3
<b>Educational status</b>								
Illiterate	12	34.3	18	60	15	93.8	45	62.7
Read and write	7	20.0	6	20	-	-	13	13.3
1-4	13	37.1	4	13.3	-	-	17	16.8
5-8	3	8.6	2	6.7	1	6.2	6	7.2
Family size (Mean±SE)	5.7±0.4 <sup>b</sup>		5.9±.6 <sup>a</sup>		5.9±.5 <sup>a</sup>		5.8±0.5	

N=Number of households SE=Standard error

**Table 2.** Species composition and livestock holdings in the study area (Mean ± SE).

Descriptor	Benatsemay	Male	Hamer	Overall
N	35	30	16	81
Cattle	6.7±0.9 <sup>c</sup>	14.1±2.1 <sup>b</sup>	29.6±6.4 <sup>a</sup>	16.8±3.1
Sheep	4.1±1.6 <sup>b</sup>	3.4±0.8 <sup>c</sup>	12.8±3.7 <sup>a</sup>	6.77±2.0
Goat	11.5±1.8 <sup>c</sup>	13±2.0 <sup>b</sup>	62.9±13 <sup>a</sup>	29.14±5.6
Chicken	17.4±2.1 <sup>a</sup>	16.7±1.4 <sup>b</sup>	15.3±1.4 <sup>c</sup>	16.4±1.6
Donkey	0.1±0.4 <sup>bc</sup>	0.7±0.2 <sup>a</sup>	0.2±0.1 <sup>bc</sup>	0.3±0.2
Bee colony	2.6±1.0 <sup>b</sup>	1.63±0.6 <sup>c</sup>	6.7±1.2 <sup>a</sup>	3.6±0.9
Total herd size	11.2±2.2	12.3±1.5	12.3±1.5	11.7±1.9

### Chicken flock structure

The average flock size of local chicken in the study area was  $13.3 \pm 0.07$  (Table 3). The highest average flock size was represented by pullets (30.79 %), followed by layers (24.05 %), cocks (17.52 %), chicks (16.64%) and cockerels (11 %). There are no significant differences ( $P < 0.05$ ) of the total flock structure among the study districts.

**Table 3.** Average local chicken flock structure of the surveyed households in the study area.

Age category	Study districts							
	Benatsemay		Male		Hamer		Overall Mean	
	Mean $\pm$ SE	%	Mean $\pm$ SE	%	Mean $\pm$ SE	%	Mean $\pm$ SE	%
Layer	$3.6 \pm 0.4^a$	27.1	$3 \pm 0.3^b$	23.0	$3 \pm 0.4^b$	22.0	$3.2 \pm 0.3$	24.1
Cock	$2.4 \pm 0.3^{ab}$	18.1	$1.9 \pm 0.3^c$	14.8	$2.7 \pm 0.4^a$	19.7	$2.3 \pm 0.3$	17.5
Pullet	$3.8 \pm 0.5^c$	28.6	$4.1 \pm 0.5^{ab}$	31.5	$4.4 \pm 0.5^a$	32.3	$4.1 \pm 0.5$	30.8
Cockerels	$1.5 \pm 0.4^a$	11.2	$1.3 \pm 0.3^{ab}$	10.0	$1.6 \pm 0.3^a$	11.9	$1.5 \pm 0.3$	11
Chicks	$2 \pm 0.5^b$	15.1	$2.7 \pm 0.7^a$	20.7	$1.9 \pm 0.6^b$	14.2	$2.2 \pm 0.6$	16.6
Total flock size	$13.3 \pm 0.4$		$13.0 \pm 0.4$		$13.6 \pm 0.4$		$13.3 \pm 0.4$	

### Chicken production system

The study area practiced extensive and semi-extensive chicken production systems. It was more of scavenging type which is supplemented with little feed. About 82.7% of the chickens are managed under a traditional or extensive chicken management system while 17.3% were using semi-extensive management system. Traditional production system was being used by 80, 83.3 and 87.5% of respondents in Benatsemay, Male and Hamer districts, respectively while 20, 16.7 and 12.5% respectively were using semi-extensive system. Most of the study districts community was sedentary.

### Role of family members

Women were more responsible (60%) for many activities like selling of chickens, feeding chickens, collecting and selling eggs, natural incubation and cleaning the chicken house in study districts. Men were

responsible for purchasing chickens and caring for sick chickens. Children also participated in various husbandry activities like feeding of chickens, harvesting egg and natural incubation and hatching egg, cleaning of bird's house, provision of supplementary feed and water.

**Table 4.** Role of family members in poultry production.

Activities	Districts											
	Benatsemay				Male				Hamer			
	Responsible bodies				Responsible bodies				Responsible bodies			
	Male < 18 years	Female < 18 years	Male ≥ 18 years	Female ≥ 18 years	Male < 18 year	Female < 18 years	Male ≥ 18 years	Female ≥ 18 years	Male < 18 years	Female < 18 years	Male ≥ 18 years	Female ≥ 18 years
Purchasing	-	-	54.3	45.7	-	-	53.3	46.7	-	-	43.7	56.3
Selling chickens	-	-	40	60	16.7	10	26.7	46.6	-	-	12.5	87.5
Caring for sick chickens	5.7	8.6	20	65.7	10	6.7	53.3	30	-	-	56.3	43.7
Feeding	5.7	11.5	5.7	77.1	23.3	26.7	20	30	12.5	18.7	6.3	62.5
Collecting	8.6	11.4	5.7	74.3	20	13.3	16.7	50	12.5	12.5	-	75
Selling egg	5.7	11.4	5.7	77.2	6.7	26.7	-	66.6	6.3	18.7	-	75
Natural incubation & hatching egg	11.4	2.9	17.1	68.6	17.1	14.3	28.6	40	12.5	6.3	37.5	43.7
Cleaning the chicken house	-	28.6	-	71.4	-	36.7	-	63.3	-	18.8	-	81.2

### Chicken housing

In Benatsemay and Male districts households keep their chickens using different types of housing systems for night sheltering while in Hamer district all households (100%) keep their chicken in the house purposely made for chicken (Table 5). The proportion of households that use a separate housing system was higher



(40%) in Benatsemay than in Male (6.7%) districts. The respondents who have no separate house kept their chicken inside the house, perch on trees (39.5%), and hand-woven basket inside the house (11.6 %). Among the interviewed households about 48.9% kept their chicken in separate house. The poultry shelters were made of corrugated iron sheet, grass/bush and wood. About 75.3% of the respondent's chicks housed with adults in the study area.

**Table 5.** Type of chicken's shelter, type of housing materials and chicken house.

Variable	Districts							
	Benatsemay		Male		Hamer		Over all total (n=81)	
	Frequency	%	Frequency	%	Frequency	%	Frequency	%
Chicken rest at night (%)								
Inside the house	10	28.	27	90.	-	-	37	39.5
Hand woven basket	11	31.	1	3.3	-	-	12	11.6
Purposely made for chicken	14	40.	2	6.7	16	100	32	48.90
Type of housing material (%)								
Iron sheet	7	20.	-	-	11	68.	18	6.7
Grass/bush	28	80.	30	10	5	31.	60	
Wood					16	100	33.3	
Chicks housed with adults (%)								
Yes	20	57.	29	96	12	75.	61	75.3
No	15	42.	1	3.	4	25	20	24.7

### Feed and water sources

Open scavenging and occasional supplementation were the major feed sources in the study area. About 93.8% of the respondents reared their chickens in an open scavenging with seasonal and regular supplementations (Table 6). The most common supplementary feed resources were maize and sorghum grains. The supplementation frequency was 44.4, 23.5, 23.5% once, twice, and three times per day respectively. Most of the respondents (74.1%) did not use feed trough, they simply pour the grain on the

ground. The results indicated that respondents discriminate classes of chickens giving supplementary feed. Layers and chicks age groups were the first and second ranked chickens receiving supplementary feed respectively.

Water is important for animals including chickens to keep them healthy and increase production. All the respondents (100%) in the study areas provided water to their chickens and tap water and river water were the major water sources.

### **Disease status**

Majority of the respondents (82.7%) in the study areas experienced disease outbreaks (Table 7). Most of the respondents in the study districts treat their sick chickens traditionally due to lack of veterinary health service and limitation of extension service. The major common disease observed in the study areas was Newcastle (53.7%), followed by Influenza (25.8%), Coccidiosis (13.4%) and Infectious coryza (7.2%). Among the identified diseases, Newcastle was economically significant infectious viral disease of chickens in the study area.

**Table 6.** Feeding practice in study area.

Variable	Districts							
	Benatsemay		Male		Hamer		Overall	
	Frequenc	%	Frequency	%	Frequency	%	Freque	%
Main feed source of chickens (%)								
Own scavenging	21	60	20	66.7	10	62.	51	63
Supplementing	14	40	10	33.3	6	37.5	30	37
Do you give supplementary feed to your chickens (%)								
Yes	33	94.3	27	90	16	100	76	93.8
No	2	5.7	3	10	-	0	5	6.2
Type of supplementary feed resources (%)								
Maize grain	19	54.3	16	53.3	11	68.7	46	56.8
Sorghum grain	11	31.4	8	26.7	2	12.5	21	25.9
Household left over	5	14.3	6	20	3	18.8	14	17.3
How frequently do you feed (%)								
Morning	6	17.2	12	40	1	6.2	19	23.5
Afternoon	3	8.6					3	3.7
Morning & Afternoon	3	8.6			1	6.2	4	4.9
Morning & evening	8	22.9	18	60	10	62.5	36	44.4
Morning, Afternoon & Evening	15	42.9	-	-	4	25	19	23.5
Feeding materials								
Containers	4	11.4	13	43.3	-	-	17	21
Ground	28	80	16	53.3	16	100	60	74.1
Containers & ground	3	8.6	1	3.3	-	-	4	4.9
Class of chickens receiving supplementary feed (index value)								
Layers	0.35		0.35		0.38		0.36	
Cock	0.21		0.10		0.04		0.12	
Pullet	0.02		0.12		0.15		0.1	
Cockerels	0.13		0.07		0.09		0.09	
Chicks	0.29		0.36		0.34		0.33	

**Table 7.** Health and disease practices in study area.

Variables	Districts							
	Benatsemay		Male		Hamer		Overall total	
	Frequency	%	Frequency	%	Frequency	%	Frequency	%
Did you experience disease outbreaks in the last 12 months?								
Yes	28	80	25	83.3	14	87.5	67	82.7
No	7	20	5	16.7	2	12.5	14	17.3
What do you do when chickens become sick?								
Treat them myself	20	57.1	29	96.7	12	75.0	61	76.3
Call in the vet.	14	40.0	1	3.3	3	18.8	18	20.7
Doctor								
Kill them immediately	1	2.9	-	-	1	6.2	2	3.0
Name of common diseases (%)								
Newcastle	18	51.4	16	53.3	9	56.3	43	53.7
Influenza	9	25.7	8	26.7	4	25.0	21	25.8
Coccidiosis	5	14.3	4	13.3	2	12.5	11	13.4
Infectious coryza	3	8.6	2	6.7	1	6.3	6	7.2

### Productive and reproductive performance

Productive and reproductive variables of indigenous chickens showed a significant difference in the studied districts (Table 8). The average age at sexual maturity of male and female was  $5.9 \pm 0.3$  and  $6.2 \pm 0.3$  months respectively. The average age at first lay was 6.7 months. The average market age of male and female were  $7.4 \pm 0.4$  and  $8.5 \pm 0.4$  months respectively. The market age was not significantly different ( $P < 0.05$ ) among study districts. The result also indicated that the average number of eggs laid in single clutch was  $13.4 \pm 0.6$  and average number of chicks hatched per incubation was  $10.5 \pm 0.4$ . The average number of chicks surviving

was  $6.1 \pm 0.3$  and number of clutches per hen per year was  $3.4 \pm 0.2$ . The higher number of clutches per year per hen was in Benatsemay ( $3.9 \pm 0.2$ ) district. The average number of eggs laid per year per hen was  $58 \pm 2.3$ .

**Table 8.** productive and reproductive performance of indigenous chickens.

Reproductive Parameters	District							
	Benatsemay		Male		Hamer		Overall Mean	
	N	Mean $\pm$ SE	N	Mean $\pm$ SE	N	Mean $\pm$ SE	N	Mean $\pm$ SE
Average age at sexual maturity of (male; month)	35	$5.5 \pm 0.1^c$	30	$6 \pm 0.2^b$	16	$6.2 \pm 0.5^a$	81	$5.9 \pm 0.3$
Average age at sexual maturity (female; month)	35	$5.8 \pm 0.2^b$	30	$6.3 \pm 0.3^{ab}$	16	$6.5 \pm 0.5^a$	81	$6.2 \pm 0.3$
Age at first egg production(month)	35	$6.4 \pm 0.1^b$	30	$6.7 \pm 0.1^{ab}$	16	$7 \pm 0.4^a$	81	$6.7 \pm 0.2$
Average market age (male, month)	35	$7.2 \pm 0.2^b$	30	$7.4 \pm 0.3^{ab}$	16	$7.5 \pm 0.6^a$	81	$7.4 \pm 0.4$
Average market age (female, month)	35	$8 \pm 0.2^c$	30	$8.5 \pm 0.3^b$	16	$9 \pm 0.6^a$	81	$8.5 \pm 0.4$
Number of chicks hatched one incubation	35	$10 \pm 0.4^b$	30	$11 \pm 0.3^a$	16	$10.4 \pm 0.5^b$	81	$10.5 \pm 0.4$
Number of chicks surviving	35	$6.0 \pm 0.3^b$	30	$6.6 \pm 0.3^a$	16	$5.8 \pm 0.2^c$	81	$6.1 \pm 0.3$
Number of eggs laid in a single clutch	35	$17.7 \pm 0.6^a$	30	$14.8 \pm 0.6^b$	16	$13.6 \pm 0.7^c$	81	$13.4 \pm 0.6$
Number of times the hen hatches in a year	35	$3.9 \pm 0.2^a$	30	$3.1 \pm 0.1^b$	16	$3.1 \pm 0.2^b$	81	$3.4 \pm 0.2$
Number of eggs produced annually	35	$64.6 \pm 3.2^a$	30	$55.8 \pm 2.3^b$	16	$53.7 \pm 1.5^c$	81	$58 \pm 2.3$

### Production constraints

The five major constraints of chicken production, in descending order of importance, were disease outbreak, predator, feed shortage, drought, and market (Table 9). Disease prevalence have been reported by the majority of respondents as common constraint and ranked first.

**Table 9.** Production constraints of indigenous chickens in study area.

Constraints	Districts												Overall index
	Benatsemay				Male				Hamer				
	Rank		Index		Rank		Index		Rank		Index		
	1	2	3		1	2	3		1	2	3		
Disease	21	4	-	0.34	15	7	-	0.33	11	-	2	0.46	0.38
Predator	10	15	4	0.30	10	13	-	0.31	5	12	-	0.35	0.32
Feed shortage	4	10	21	0.25	5	-	20	0.19	-	4	10	0.17	0.20
Drought	-	6	10	0.10	-	10	-	0.11	-	-	4	0.03	0.08
Market	-	-	-	-	-	-	10	0.06	-	-	-	-	0.02

## DISCUSSION

Compared to earlier studies conducted on poultry production in the country, some differences were observed in the current study area. Most of the households in this study were male headed which is lower than the report by Fitsum et al. (2017) in central zone of Tigray region in northern Ethiopia. There was a difference between districts in educational level. The level of illiterate was highest in Hamer district. In contrast to this finding, better education level was reported from Southern Ethiopia including lower proportions of illiterate and higher number of people with reading and writing ability (Melak et al., 2021). Thus, better educational background obtained in Benatsemay and Male districts might be a good potential for conservation and sustainable utilization of chickens. It is also be useful to consider upgrading the education status in Hamer district for successful chicken breeding strategies and sustainable utilization interventions.

The average family size of the households was closer to the report from Jimma and Illu Aba Bora zones, southwestern Ethiopia (Haile and Biratu, 2017). However, the family size of all districts in this study was higher than the average value of Ethiopia (CACC, 2003).

The average flock size per household was higher than the reported size in Sheka zone (Assefa et al., 2019), Kambata Tambaro and Wolita Zones (Getiso et al., 2015), Northwest Ethiopia (Halima et al., 2007) and South Ethiopia (Mekonen, 2007) and similar with what has been reported from North Gondar Zone, and Ethiopia (Getu and Birhan, 2014). Compared to other countries, the flock size per household was lower than that of Jordan (Abdelqader et al., 2007) and Pakistan (Hunduma et al., 2010).

The flock owner of the chicken determines the flock composition based on economic and management considerations. The number of local chickens in the household in different age categories varies considerably. On average pullets followed by layers were dominant in in the present study area. Which is in contrast to the findings from Northern Gonder, Ethiopia (Wondu et al., 2013). The higher proportion of pullets in the study districts indicated the measures that has been taken to get replacement flocks of layers for egg production and chicken production. This would have direct impact on conservation and sustainable utilization of the resource.

The current result showed that the dominant chicken production system was traditional or extensive type. This agrees with the findings of South west and South part of Ethiopia (Moreda et al, 2013). All members of the family were responsible for poultry activities. This finding was similar with Jamma woreda, south Wollo (Mammo, 2006) and Ganta Afeshum district of Eastern Tigray, Ethiopia (Gebresilassie et al., 2015). Participation of all family members in poultry activities might suggest that poultry keeping is an unbiased practice which allows income generation and sharing of benefits among family members.

About 48.9% of the households kept their chicken in separate house. This finding is higher than what has been reported from GantaAfeshum district of Eastern Tigray (Gebresilassie et al., 2015), North West Ethiopia (Halima, 2007) and Jamma woreda, south Wollo (Mammo, 2006). In Hamer district all respondents

used separate house for chicken. This showed that in Hamer district the owners are aware of the importance of providing separate house for chickens. The differences observed among the study districts might be due to lack of awareness on the importance of chicken house in Benatsemay and Male districts. Locally available materials were used for constructing chicken shelters similar to the reports Gebresilassie et al. (2015) and Halima et al. (2007).

The major feed sources in the study districts were scavenging with occasional supplementation and the major water sources were tap and river water. These results were similar to that of Fitsum et al. (2017). The supplementation frequency of the study area is in line with that reported in Pawe District, Beneshangul Gumuz region, Ethiopia (Dejene, 2021).

Newcastle disease was the most common and economically significant infectious viral disease of chickens in the study area. The result was similar with Serkalem et al. (2005) and Gebremedhin (2007) who reported that this disease was the major infectious diseases affecting productivity and survival of village chicken in the central highlands of Ethiopia. For conservation and sustainable utilization strategies, chicken producers should be encouraged to adopt proper Newcastle and other disease's control measures and the limited animal health services need to be strengthened.

The average age at sexual maturity of male and female was almost similar with those reported in Sheka zone, south western Ethiopia (Assefa et al., 2019) and in Dawro zone and Konta special district, southern Ethiopia (Melak et al., 2021). The average age at first egg laying was higher than the findings of Fitsum et al. (2017). The clutch number of chickens in the study area was similar with the reports of Matawork et al. (2019) in Gena Bossa district of Dawro Zone, Ethiopia and Meseret (2010) in Gomma district, but lower than the clutch numbers reported in Bure and Dale districts, respectively (Fisseha et al., 2010).

The survival rate in the present study was lower than the one reported by Fisseha et al. (2010). The low survival rate might be due to prevalence of diseases, predators and lack of vaccination practice in the study area. The average number of eggs per year per hen was higher compared to the results identified in earlier



studies (Assefa et al., 2019; Markos et al., 2015; Addisu, 2013; Ayalew and Adane, 2013; Meseret, 2010; Halima et al., 2007) but lower than the reports by Fitsum et al. (2017), Fisseha et al. (2010) and Mekonnen (2007).

The Disease prevalence was the most challenging constraints in the study area. This result was similar with report from southern Ethiopia (Melak et al., 2021). This might be due to the lack of healthcare services in the study area.

### **CONCLUSION AND RECOMMENDATION**

The poultry production system in the study area was more of a traditional production system. The major production constraints were disease, predator and feed shortage. Indigenous chicken populations have potential for egg and meat production and the reproductive performances are reasonable under the existing limiting environmental factors. The type, seasonal occurrence and economic loss due to diseases, predator and feed shortage should be documented and pertinent control measure need to be introduced. The constraints of indigenous chicken production can justify for the need of appropriate community-based conservation and sustainable utilization strategies so as to conserve the genetic resource as well as benefitting the community.

### **ACKNOWLEDGEMENTS**

The authors would like to sincerely thank to all smallholder farmers and pastoralists in the study area who allowed their time for interview. We are also grateful to the Ethiopian Biodiversity Institute for the provision of the research fund.

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## Factors Influencing Climate-Smart Agriculture Practices Adoption and Crop Productivity among Smallholder Farmers in Nyimba District, Zambia

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**ABSTRACT:** Many smallholder farmers in the developing world live in adverse poverty and rely on agriculture as their primary source of income and household food. This study examines factors influencing the adoption of climate-smart agriculture practices and crop productivity among smallholder farmers in Nyimba District, Zambia. Data were collected from June to July of 2022 from 194 smallholder farmers' households in twelve villages belonging to four agricultural camps of Nyimba District. Four focus group discussions were conducted to supplement data collected from the household interviews. A binary logistic regression model was used to assess the determinants of climate-smart agriculture adoption and crop productivity among smallholder farmers. Propensity score matching was performed to measure the impacts of climate-smart agriculture adoption among adopters and non-adopter farming households. The Logistic regression model showed that the smallholder farmer's level of education, household size, synthetic fertilizer usage, age of household head, gender, farming experience, livestock ownership, annual income, farm size, marital status of household head, and access to climate information, all affect smallholder farmer climate-smart agriculture practices adoption and crop productivity. The propensity score matching the analysis found overall crop yield (for entire crops) was 20.20% higher for climate-smart agriculture practices adopters than for non-adopters. The study also found smallholder farmers' climate-smart agriculture practices adopters maize yield (staple crop) increased by 21.50% higher than non-adopters. The findings from this study have implications for further research and policy design and implementation of climate-smart agricultural practices.

**Keywords:** Adoption, Agriculture, Climate-smart agriculture, Climate change, Crop productivity.

## INTRODUCTION

Climate changes are already hampering agricultural production growth for both livestock and crop production worldwide (Alfani et al., 2019). Increased climate variability and climate change exacerbate production risks and challenge farmers' coping abilities. These climate changes bring about threats to access nutritious food for urban, peri-urban, and rural communities due to reduced agricultural production and household income (Ivanova et al., 2020; Sharifi, 2021; Mossie, 2022), and increased risks that disrupt food markets. According to the Intergovernmental Panel on Climate Change (IPCC) 2018 report, climate change affects crop production in most parts of the world, with negative effects more common than positive, and developing countries remain extremely susceptible to further negative impacts. Increases in the frequency and intensity of extreme events such as drought, heavy rainfall, flooding, and high maximum temperatures are already occurring and are expected to accelerate in many parts of the world (Murray and Ebi, 2012; IPCC, 2018). Average and seasonal maximum temperatures are projected to continue rising with higher average rainfall overall. These effects will not, however, be evenly distributed and are likely to increase by the end of the 21<sup>st</sup> century.

Climate change is projected to partake in and contribute to a worldwide reduction in cereal yields (*i.e.*, maize and wheat by 3.8% and 5.5% respectively (Lobell et al., 2011). Smallholder farmers falling in the group of poor producers, the landless, and marginalized ethnic, are all vulnerable to changes in climate (CIAT and World Bank, 2017; Makate, 2019). In addition, climate change extreme events and shocks can be long-lasting, as risk exposure and increased uncertainty affect investment incentives and reduce the likelihood of effective farm innovation while increasing that of low-risk, low-return activities. Climate change will almost certainly have a significant impact on the average yields of Zambia's major crops (maize, wheat, and sorghum), because agronomic conditions for these crops may worsen in large parts of the country

(Molieleng et al., 2021; Chavula, 2022). Climate change extreme events and shocks such as drought and flooding, do have a greater impact on crop production in Zambia and other Sub-Saharan African countries. However, through the intricacy of the agricultural diverse systems in Sub-Saharan African countries and its interrelation between the socio-economic facets of smallholder farmers' households, an integrated approach has been promoted to sustainably increase the productivity of smallholder agricultural landscape to adapt to climate change. These approaches and/or interventions are termed 'climate-smart agriculture (CSA)' farmers (Makate, 2019; Odubote and Ajayi, 2020; Zakaria et al., 2020; Molieleng et al., 2021). Climate-smart agriculture practices (e.g. sustainable agriculture, integrated nutrient management, organic farming, agroforestry technologies, integrated pest management, conservation agriculture, multi-cropping system, among others) are designed to increase household income, improve agricultural production while promoting climate change resilience through sustainable management of arable land and less synthetic fertilizer usage (Newell et al., 2019).

Climate-smart agriculture emerged in the late twentieth century in Zambia, when the country began facing economic, ecological, and/or climate change challenges in line with their agriculture production. The emergency of CSA focused on combating the adverse impacts of climate change on smallholder farming households, the country has embarked on the promotion of CSA practices to reclaim degraded landscapes and enhance households' resilience to climate change (Ngoma et al., 2021). Subsequently, due to the importance of CSA, the country has made climate-smart agriculture practices' promotion (*i.e.*, organic farming, integrated pest management, agroforestry, conservation agriculture, and integrated agriculture practices to mention a few) among the most important components of extension and rural advisory service delivery. These interventions have been conducted in concurrence with national and international research, non-governmental organizations, and development partners (Ngoma et al., 2021). Several studies in Zambia have been conducted to investigate the impact of CSA on smallholder farmers' livelihoods, especially those living in rural areas. Most of these studies have focused on the impacts of CSA practices' adoption on

smallholder farmers' household income as a measure of adopters' household livelihood (Kalaba et al., 2010; Kuntashula and Mungatana, 2015; Jama et al., 2019; Nkhuwa et al., 2020). Nkhuwa et al. (2020) and Kuntashula and Mungatana (2015) found that implementing improved fallow and green leaf manure as agroforestry practices considerably boosted smallholder farmers' household income. Jama et al. (2019) observed agroforestry adoption enhanced household income by improving fallow adoption by smallholder cotton growers and Kalaba et al. (2010) revealed that adopting agroforestry practices improved smallholder farmers' household welfare in Southern African nations including Zambia. In Zambia, there appears to be scanty information related to factors influencing climate-smart agricultural practices, adoption, and crop productivity among smallholder farmers in Nyimba district. Hence, this study, unlike earlier empirical studies, examines the factors influencing climate-smart agricultural practices, adoption, and crop production among smallholder farmers in Nyimba district, Zambia.

### **Conceptual Framework**

Climate-smart agriculture is a strategy for changing and reorienting the agricultural landscape to promote food security in light of the emerging climatic realities, variations, and climate change (Chavula, 2021). Climate change disrupts food markets, posing population-wide risks to food production and supply. These risks can be decreased by enhancing farmers' capacity for adaptation as well as enhancing the mitigation and efficiency of agricultural production systems. Smallholder farmers who have received information on climate change and/or perceive it to be real are highly likely to adopt climate-smart agricultural practices to meet its tenets to boost household income and productivity; increase resilience and adaptation; mitigate and reduce greenhouse gas emissions. The adoption of climate-smart agriculture to meet its tenets is affected by institutional, cognitive, and socio-economic factors (Annex 1).

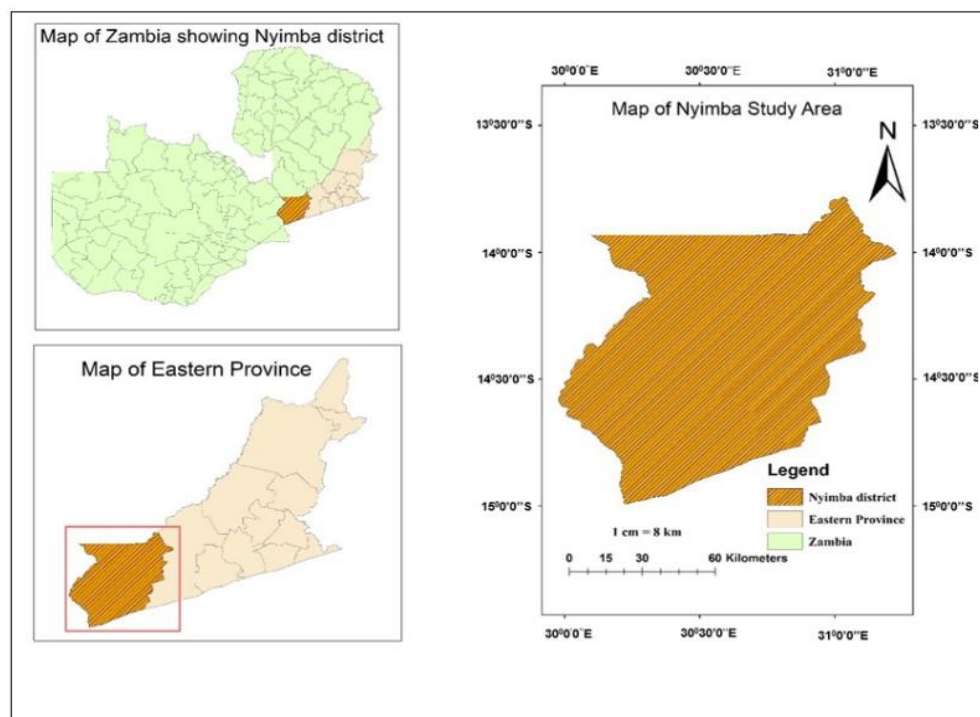


## MATERIALS AND METHODS

### Study area description

#### Location

The research was carried out in the Nyimba district of Eastern Province, Zambia. The district is situated 334 kilometers east of Lusaka Zambia's national capital. In the South the district borders with Mozambique, North with Muchinga province, West with Lusaka province, and East with Petauke district. The district lies between latitude ( $13^{\circ}30'1019''$  and  $15^{\circ}55'8146''$  South) and longitude ( $30^{\circ}48'5047''$  and  $31^{\circ}48'20252''$  East) (Figure 1).



**Figure 1.** Map of the study area.

#### Climate, soil, and topography

Zambia as a country is divided into three agro-ecological zones (*i.e.*, Zone I, Zone II (IIa and IIb), and Zone III) of which Nyimba district falls in Zone I. Agro-ecological zone I covers the Zambezi and Luangwa River basins' Southern and Eastern rift valleys. It also stretches to parts of Zambia's Western and Southern provinces in the south (Mtambo et al., 2007). The district's average annual rainfall ranges between 600 to

900 millimeters; the wettest months are December to February, with a distinct dry season from May to November. The annual mean temperature is 24.2°C whereas the daily temperature range is 10.3 °C to 36.5 °C. Topographically, the district is composed of hills and plateaus, soils characterized as Lithosol-Cambisols, whereas in the valleys, soils are classified as Fluvisol-Vertisols. The elevation varies from 450-1000m at the Luangwa River valley bottom and extends to the plateau near Nyimba district center, and even higher on the mountain tops in the district's western part (Halperin et al., 2016).

### **Vegetation type**

The Miombo woodland is the most dominant formation and habitat type in Southern Africa (Gumbo and Dumas-Johansen, 2021; Montfort et al., 2021). Miombo woodland is also the major forest type in Zambia itself, covering approximately 45% of the entire land surface (Kalinda, 2008). Nyimba is located in the middle of the Miombo Ecoregion, a biome with a variety of flora types that is dominated by tree species from the Caesalpinioideae subfamily of leguminous plants (Timberlake and Chidumayo, 2011). Depending on the climate, soil, landscape position, and degree of disturbance, the ecoregion's vegetation varies in composition and structure (Timberlake and Chidumayo, 2011; Halperin et al., 2016). Nyimba is located in the arid ecozone and is characterized by four types of vegetation: Dry miombo woodland (*i.e.*, *Brachystegia spiciformis*, *B. boehmii* and *Julbernardia globiflora*), Mopane woodland (*i.e.*, *Colophospermum mopane*), Munga woodland (*i.e.*, *Vechellia* sp., *Senegalia* sp., *Combretum* sp., and trees associated with the Papilionoideae subfamily) and Riparian Forest (*i.e.*, mixed tree species).

### **Land use and farming systems**

Nyimba district's total land area is about 10,500 square kilometers according to the population and housing census of 2010 (Central Statistical Office, 2011). Therefore, 82% of the district population is agrarian and three-quarters are impoverished, living in rural areas, and earning less than the international poverty threshold of \$2.15 a day. These households are farmers who are into mixed agriculture practices dominating the district. Under this agricultural system, crops are grown in mounds or ridges, in most cases maize. The

major crops grown include banana (*Musa* sp.), maize (*Zea mays*), finger millet (*Eleusine coracana*), groundnuts (*Arachis hypogaea*), haricot bean (*Phaseolus vulgaris*), cowpeas (*Vigna unguiculata* spp.) and soybean (*Glycine max*). Multiple cropping systems are common where the cultivated land is on gently and moderately steep slopes. The topography of the land in the district makes the agricultural cultivation pattern different from other areas. Therein, the cropping system is alongside livestock production such as cattle, goats, chickens, ducks, and doves. Besides agricultural activities, farmers are engaged in charcoal production, timber, firewood supply, and non-timber forest products (NTFPs) from the miombo woodland for household economic gain (Gumbo et al., 2016).

### **Site selection**

The selection of the study area was based on non-governmental organizations implementing CSA projects in Nyimba district. Non-governmental organizations for over 15 years and currently work with 80 community cooperatives providing relevant farmer support services to more than 69,000 farmers' households. These organizations are well embedded with local communities and have long experience working on CSA intensification through networks of peer-selected lead farmers to maximize outreach and knowledge sharing. This existing system enabled the study to conduct a reconnaissance to gather basic information about the study area before data collection. Information gathered included; distance between villages, number of farming households per village, contact details for lead farmers, CSA practices of adopters' households, and the location of croplands, and identifying central meeting points for focus group discussion (FGD).

### **Data sources**

This research employs both quantitative and qualitative data collection techniques and both primary and secondary sources as data sources. The primary data sources for this study were obtained through a structured questionnaire and crucial oral interviews with sample households and key respondents. The Agricultural Office, extension officers, lead farmers, project reports and paperwork, further research papers,

demographic and socioeconomic profiles, and published materials such as books and journals were used as secondary sources for this study.

### Sampling technique

This research used a multistage random sampling technique to select participants to be part of the study. This study drew smallholder farmers from agricultural camps. An agricultural camp is a delineation made by the Republic of Zambia Ministry of Agriculture containing a certain number of smallholder farmers' households in a district across villages for easy access by agriculture extension officers. From the eight agricultural camps in Nyimba District, four agricultural camps were randomly selected (*i.e.*, Ndake, Central camp, Lwende, and Ofumaya). The total number of farmers in the selected four agricultural camps in Nyimba District is 10,700. The study used Slovin's formula for sample size calculation. Furthermore, the study randomly selected three villages from each camp (*i.e.*, Sikwenda, Sichipale, Mawanda, Elina, Katumbila, Sichalika, Malalo, Mwenecisango, Mulivi, Lengwe, Mofu and Yona). The study first used a margin of error of 0.05 and obtained a sample size of 386 participants. However, as this sample size required more time and resources, to reduce the sample size, the study then used a margin of error of 0.1 and obtained a size of 99, as shown below.

Sample size formula: Slovin's (1960) formula:

$$n = \frac{N}{1 + Ne^2}$$

$$n = 10700 / (1 + 10700(0.1^2))$$

$$n = 10700 / 27.75 \quad n = 99.07$$

The study therefore settled for a sample size of 194 participants, which is between the sample size of 99 (0.1 margin of error) and 386 (0.05 margin of error). Through the aid of agricultural camp officers, farmer registers for each village were used to randomly select participants in an Excel spreadsheet.

**Focus group discussion**

Focused group discussions (FGD) were conducted to collect in-depth data about smallholder farmers' factors affecting climate-smart agriculture practices (CSAP) adoption, and crop productivity. This was attained through means of a developed open-ended FGD study tool. The FGDs are regarded to be better than individual interviews as sensitive issues come out during the implementation. A total of four (FGDs) were carried out in the study area comprising village headmen, women, men, and youths. The FGD meetings were held at central places for easy access by individual farmers.

**Household interviews**

A household survey was utilized to obtain quantitative and qualitative data from the sampled smallholder farmers in the study area. To obtain data, a semi-structured questionnaire comprising open-ended and closed questions was employed. However, data on the socioeconomic, institutional, and demographic characteristics of the sampled homes were attained from smallholder farmers' households. Before beginning the data collection activity, the questionnaires were pretested multiple times for suitability (e.g., clarity, adequacy, and question sequence), correctness, and coherence of the survey questions, and the findings were used to make changes. The questionnaire was pretested on 23 randomly selected households that were not part of the survey's sampled group. The researcher trained enumerators after pretesting and before presenting questionnaires to smallholder farmers on the final interview schedule. Finally, the enumerators gathered information under the supervision of researchers and supervisors. Collected data was verified and amended after each fieldwork day and backed to CSPRO Cloud.

**Data quality control**

Before performing data analysis, the household survey data was scrutinized on six dimensions: (1) correctness, (2) completeness, (3) consistency, (4) timeliness, (5) validity, and (6) originality. As a result, duplicated data, incomplete data, inconsistent data, poorly organized data, and inadequate data were eliminated.

**Data analysis**

Data from the household survey was analyzed with STATA 15MP for descriptive statistics such as mean, frequency, standard deviation, and percentage to describe household characteristics and socio-economic dynamics among CSA practices, adopters, and non-adopters smallholder households.

**Variables specification****Outcome variables**

The outcome variable for this study is the impact of CSA practice adoption among smallholder farmers' households' crop productivity.

**Dependent variables****Smallholder farmers' household decision to adopt CSAPs**

The dependent variable was the smallholder farmers' household to adopt CSAPs taking a value of one (1) and zero (0) if the smallholder farmers' household does not adopt. The main reason was to identify factors that influence the adoption of CSAP among smallholder farmers' households in the Nyimba district, Zambia.

**Propensity score matching**

Propensity score matching (PSM) method was used in this study to determine the effect of CSAP on crop productivity among adopters and non-adopters. Propensity score matching is a way of correcting treatment effect estimates by adjusting for confounding variables across a sampled population. According to Caliendo and Kopeinig (2008), there are steps in implementing PSM for a study. These are estimation of the propensity scores using a binary model, choosing a matching algorithm, checking against a common support condition, and testing the matching quality of the treatment and/or participants (Caliendo and Kopeinig, 2008).

**Step 1: Model Specification**

The Logit model in this research can be preferred due to the consistency of parameter estimation associated with the assumption that the error term in the equation has a logistic distribution (Baker, 2000; Ravallion, 2001). Therefore, the Logit model was used to estimate the probability of smallholder farmers' adoption of CSAPs allotted to socio-economic, agroecological, and institutional characteristics. Therein, a dependent variable is considered a value of 1 for CSAP adoption and 0 for non-CSAP adopters.

$$P_i = P(Y = 1|X) \quad (1)$$

In line with Pindyck and Rubinfeld (1981), the cumulative logistic probability function is specified as follows;

$$P_i = F(Z_i) = F\left[\alpha + \sum_{i=1}^m \beta_i X_i\right] = \left[\frac{1}{1+e^{-(\alpha+\sum \beta_i X_i)}}\right] \quad (2)$$

where  $e$  represents the base of natural logs,  $X_i$  represents the  $i^{\text{th}}$  explanatory variable,  $P_i$  is the probability that a household adopts CSAPs, and  $\alpha$ , and  $\beta_i$  are the parameters to be estimated.

Interpretation of coefficients is made easier if the logistic model can be written in terms of the odds and log of odds (Gujarati, 1995). The odds ratio implies the ratio of the probability that an individual will be a participant ( $P_i$ ) to the probability that he/she will not be a participant ( $1-P_i$ ). The probability that he/she will not be a participant is defined by:

$$(1 - P_i) = \frac{1}{1+e^{zi}} \quad (3)$$

$$\left(\frac{P_i}{1+P_i}\right) = \left[\frac{1+e^{zi}}{1+e^{-zi}}\right] = e^{zi} \quad (4)$$

Alternatively,

$$\left(\frac{P_i}{1+P_i}\right) = \left[\frac{1+e^{zi}}{1+e^{-zi}}\right] = e^{[a+\sum B_i X_i]} \quad (5)$$

Taking the natural logarithms of equation (3.5) will give the logit model as indicated below.

$$Z_i = \ln\left(\frac{P_i}{1-P_i}\right) = a + B_1X_{1i} + B_2X_{2i} + \dots + B_mX_{mi} \quad (6)$$

If consider a disturbance term,  $\mu_i$ , the logit model becomes

$$Z_i = a + \sum_{t=1}^m B_t X_{ti} + \mu_i$$

So the binary logit will become:

$$Pr(pp) = f(X) \quad (7)$$

Where  $pp$  is CSAPs adoption,  $f(X)$  is the dependent variable project adoption, and  $X$  is a vector of observable covariates of the households. The dependent variable will take a value of 1 for CSAP adoption and 0 for non-adopters.

In addition to the estimated coefficients, the marginal effects of the change in the explanatory variables on the probability of CSAP adoption are also estimated. The interpretation of these marginal values will be dependent on the unit of measurement for the explanatory variables. However, when the explanatory variable is dummy, the marginal effects generally produce a reasonable approximation to the change in the probability that  $Y = 1$  at a point such as the regressors' average.

## **Step 2: Defining the Region of Common Support and Balancing Tests**

The region of common support needs to be defined where distributions of the propensity score for treatment and comparison groups overlap. Sampling bias may still occur, however, if the dropped CSAP's non-adopters observations are systematically different in terms of observed characteristics from the retained non-adopters; these differences should be monitored carefully to help interpret the treatment effect. Balancing tests can also be conducted to check whether, within each quantile of the distribution of propensity scores, the average propensity score and mean of  $X$  are the same. For PSM to work, the comparison and treatment groups must be balanced in that similar propensity scores are based on similar observed  $X$ . The distributions



of the treated group and the comparator must be similar, which is what the balance implies. Formally, one needs to check if  $\hat{P}(X|T = 1) = \hat{P}(X|T = 0)$ .

### **Step 3: Matching Adopters to Non-adopters**

The third step is to choose an algorithm for data matching available. Matching is a common method for deciding on control subjects who are matched to the treated subjects based on context covariates that the investigator believes need to be monitored. Different ones may employ matching standards. to assign adopters to non-adopters based on the propensity score. The most common matching algorithms are nearest neighbor matching (NN), radius matching (RM), and kernel-based matching (KBM).

### **Step 4: Matching Quality**

In the fourth step, matching quality tests could be done. Checking for matching regardless of quality, the matching methods can balance the distribution of various variables or not. If differences exist, there may be an indication of incomplete matching, and remedial actions are suggested (Caliendo and Kopeinig, 2008). The following step is to check whether the treatment introduced a distinction in the indicators of impact. The average treatment effect of the treated (ATT) is given by the distinction within the mean outcome of matched adopters and nonadopters that have common support conditional on the propensity score.

### **Step 5: Sensitivity Analysis**

Finally, a sensitivity analysis will be carried out to check the conditional independence assumption strength. Sensitivity analysis also will be utilized to look at whether an unmeasured variable's effect on the choice process is strong enough to jeopardize the matching approach (Ali and Abdulai, 2010). The Rosenbaum bound sensitivity test will be used to carry out the sensitivity analysis (r-bounded test).

## **RESULTS**

### **Characteristics of the participant smallholder farmers**

The household survey comprised 194 smallholder farmer participants from the research area, who were chosen at random. The smallholder farmers were interviewed about crop production and their applications

of various CSA practices. The study presents the household survey's findings, starting with the demographic characteristics of the participants, crop production and productivity, adoption of CSA, constraints on the adoption of CSA practices, effects of CSA practices on crop productivity, and factors affecting crop productivity. The study obtained a total of 339 field plots of various crops from 194 farmer participants. From the results in Table 1, the study obtained that the mean age of the respondents was 46 years of age, with a standard deviation of 14.59. A majority (62.18%) were male-headed households, and 69.43% were married. The mean year of formal education was found to be 5.49 years, with a standard deviation of 3.5. The mean year of farming was found to be 26.22, with a standard deviation of 15.55. Concerning the years of living in the area, the mean was 30.92, and the standard deviation was 18.68. The average family size was 5.42, with a standard deviation of 2.14. The average total annual income was revealed to be K 5472.68 (USD 331.68) (K 16.5 per 1 USD), and 57.51% reported participating in any off-farm activities. Improved seed varieties were used by 78.76% of the smallholder farmer participants. The average farm size (landholding) was 3.396 ha, with a standard deviation of 3.363. The land tenure system was all customary land (100%). The mean cultivated land was 1.83 ha and 1.45 standard deviation. The average number of crops grown by smallholder farmers was 2, with a standard deviation of 0.930.

**Table 1.** Characteristics of the participant smallholder farmers.

Variable	Mean	Std. Deviation
HH Head Age	46.181	14.593
HH Head Sex	Male: 62.18% (120)	
Marital Status	Married: 69.43% (134)	
Years of formal education	5.487	3.499
Years of farming	26.218	15.545
Years of living in the area	30.917	18.680
Household size	5.420	2.137
Total Annual Income (in Kwacha)	5472.689	7626.52
Participation in any off-farm activity	Yes: 57.51% (111)	
Used Improved Maize Seed	Yes: 78.76% (152)	
Farm Size (ha)	3.396	3.363
Land tenure system (Customary)	100% (194)	
Cultivated land (2021/2022), ha	1.828	1.448
Number of Crops (2021/2022)	2	0.930

HH - household

### Crops grown by smallholder farmers

Concerning the crops grown by the farmers, the study found that maize ranked first, reported in 194 crop plots, followed by groundnuts, reported in 99 plots, sunflower in 69 plots, and soya beans in 16 plots (Table 2). Other crops; Cowpea, Bambara nuts, Cotton, Millet, and Sweet Potatoes were reported to have been grown in a few plots.

**Table 2.** Crops grown by smallholder farmers.

Crops Grown	Frequency	Percent	Cumulative
Maize	194	50.13	50.13
Soybeans	16	4.13	54.26
Groundnuts	99	25.58	79.84
Cowpea	2	0.52	80.36
Bambara nuts	2	0.52	80.88
Sunflower	69	17.83	98.71
Cotton	1	0.26	98.97
Sweet potatoes	3	0.78	99.74
Millet	1	0.26	100
<b>Total</b>	<b>387</b>	<b>100</b>	

### Climate-smart agriculture practices adopted by smallholder farmers

From the results obtained, pot-holing (basin) was implemented in 61 field plots (17.99%), multi-cropping in 50 plots (14.75%), minimum tillage in 34 plots (10.03%), ripping in 32 plots (9.44%), crop rotation in 18 plots (5.31%), and manure in 11 plots (3.24%) as well as alley cropping in 9 plots (2.65%) (Table 3). The other CSA practices were implemented in a few plots less than ten.

**Table 3.** Climate-smart agriculture practices adopted by smallholder farmers.

CSA Practices	Frequency	Percent
Ripping	32	9.44
Basin	61	17.99
Crop rotation	18	5.31
Crop residue	2	0.59
Alley cropping	9	2.65
Multi cropping	50	14.75
Contour ploughing	6	1.77
Compost	5	1.47
Manure field	11	3.24
Zero tillage	34	10.03
Bunding	2	0.59

### Number of climate-smart agriculture practices adopted by smallholder farmers

Concerning the number of CSA practices adopted, no single CSA practice was implemented in 167 plots (49.26%), one CSA practice was implemented in 123 plots (36.28%), two CSA practices were implemented in 43 plots (12.68%), 4 plots had three different CSA practices implemented, and only 1 plot had four CSA practices implemented and another plot with five CSA practices implemented (Table 4). Based on these results, farmers' implementation of many CSA practices in a single plot was found to be very low.

**Table 4.** Number of climate-smart agriculture practices adopted by smallholder farmers.

No. CSA Adopted/Plot	Freq.	Percent	Cum.
0	167	49.26	49.26
1	123	36.28	85.55
2	43	12.68	98.23
3	4	1.18	99.41
4	1	0.29	99.71
5	1	0.29	100
<b>Total</b>	<b>339</b>	<b>100</b>	

### Quantities harvested for various crops (kg)

Maize, groundnuts, sunflower, and soya beans were the most grown crops by the farmers (Table 5). The mean quantity of harvest for all crops was 1223.51 kg with a standard deviation of 1442.82. The mean quantity of maize harvested for maize was 1766.57 kg with a standard deviation of 1594.23, while the mean quantity of groundnuts harvested was 511.08 kg with a standard deviation of 605.07, the mean quantity of 609.67 kg with a standard deviation of 513.02 for sunflower, while for soya beans the mean quantity harvested was 1007.5 kg with standard deviation of 1835.615.

**Table 5.** Quantities harvested for various crops (kg).

Variable	Obs	Mean	Std. Dev.	Min	Max
All Crops	339	1223.51	1442.82	50	9450
Maize	173	1766.57	1594.23	165	9450
Groundnuts	85	511.08	605.07	50	3450
Sunflower	61	609.6721	513.0212	50	2800
Soya beans	14	1007.5	1835.615	200	7245

### Productivity of various crops (Yield (Kg) per hectare)

Concerning the productivity of various crops, the overall yield per hectare of all crops was 1316.60 kg. The yield per hectare for maize was found to be at 1682.52 kg per hectare, and for groundnuts, Sunflower, and soybean the mean yield per hectare was found to be 822.90 kg, 962.79 kg and 808.40 respectively.

### Impact of climate-smart practices on crop productivity among smallholder farmers

The study investigated how climate-smart agriculture techniques affected smallholder farmers' crop yield. The study found that crop yield for CSA adopters was 20.20% higher than the CSA non-adopters (Table 6). The results were statistically significant at 0.027 p-value ( $p < 0.05$ ). This entails that adopting CSA practices increases crop yield.

**Table 6.** Impact of climate-smart practices on crop productivity among smallholder farmers.

Treatment-effects estimation		Number of Obs = 194				
Estimator: propensity-score matching		Matches: requested = 1				
Outcome model: matching		min = 1				
Treatment model: logit		max = 2				
log_yield	Coef.	AI Robust Std. Err.	Z	P>z	[95% Conf.	Interval]
ATE						
CSA_Practice						
(Adopters						
vs						
Non_Adopters)	.2019652	.0911943	2.21	0.027**	.0232276	.3807028

\*\*\*<1%, \*\*<5% and \*<10%

### Impact of climate-smart practices on maize productivity among smallholder farmers

The study conducted a propensity score matching analysis to specifically determine how CSA affects maize productivity (Table 7). The research showed that implementing CSA increases maize yield for adopters by 21.50% higher than non-adopters. This shows that adopting CSA practices significantly increases maize yield. The results were statistically significant at 0.035 p-value ( $p < 0.05$ ).

**Table 7.** Impact of climate-smart practices on maize productivity among smallholder farmers.

Treatment-effects estimation					Number of Obs = 194	
Estimator: propensity-score matching					Matches: requested = 1	
Outcome model: matching					min = 1	
Treatment model: logit					max = 1	
log_yield	Coef.	AI Robust Std. Err.	Z	P>z	[95% Conf.	Interval]
ATE						
CSA_Practice						
(Adopters						
vs						
Non_Adopters)	0.215012	0.101795	2.11	0.035**	0.015496	0.414527

\*\*\*<1%, \*\*<5% and \*<10%

### Factors affecting smallholder farmers' adoption of climate-smart agricultural

The study conducted a logistic regression analysis to determine factors affecting the adoption of CSA practices. Age has a favorable impact on the adoption of CSA practices, the higher the age, the more likely a farmer will adopt CSA practice, statistically significant ( $p < 0.001$ ). The study recorded the age category of 40-55 years and > 55 years to have adopted more CSAP in the study area.

Adopting CSA practices is influenced by farming experience, the more years a farmer spends in farming, the less likely a farmer will use CSA practices, statistically significant at 0.0000 p-value ( $p < 0.001$ ). Income was found to have a statistically positive effect on the adoption of CSA practices, the greater a farmer's income level, a farmer is more likely to adopt CSA practice, statistically significant at 0.0640 p-value ( $p < 0.1$ ) (Table 8). On the other hand, the size of the farm, the distance between the farmers' homes and the farm sites, the location, and the rise in temperature all harmed the farmers' intention of technology adoption. Gender was found to have statistically significant effect at 0.0660 p-value ( $p < 0.1$ ). Farm size was also found to have a negative significant effect on climate-smart agricultural practices adoption at 0.0050 ( $p < 0.01$ ). Livestock quantity was also found to have a significant effect on CSA adoption at 0.0180 p-value ( $p < 0.1$ ), while access to climate information had a negative influence on climate-smart agriculture adoption p-value

0.0060 ( $p < 0.01$ ). On the other hand; marital status, education, fertilizer, credit access, and access to extension services were found not to have a significant effect on the adoption of CSA practices.

**Table 8.** Factors affecting smallholder farmers' adoption of climate-smart agricultural practices.

Logistic regression				Number of Obs = 194		
Log pSeudolikelihood = -204.0124				Wald chi2(10) =	27.34	
				Prob > chi2 =	0.0112	
				Pseudo R2 =	0.0965	
CSA_Practice	Coef.	Robust Std. Err.	z	P>z	[95% Conf. Interval]	
Age	0.085697***	0.0222	3.8600	0.0000	0.0422	0.1292
Gender	0.017260*	0.4056	0.4400	0.0660	0.7776	0.8122
Marital_status	-0.178756	0.1399	-1.2800	0.2010	-0.4530	0.0955
Education	-0.051048	0.0387	-1.3200	0.1870	-0.1270	0.0249
Farming_experience	0.087116***	0.0200	-4.3600	0.0000	-0.1263	-0.0480
Household_size	-0.027906	0.0658	-0.4200	0.6720	-0.1569	0.1011
Income	0.000035*	0.0000	1.8500	0.0640	0.0000	0.0001
Fertilizer	0.000727	0.0007	1.1200	0.2630	-0.0005	0.0020
Farm_size	-0.02006**	0.0449	-0.4500	0.0050	-0.1082	0.0680
Livestockqt	0.006734*	0.0083	0.8100	0.0180	-0.0230	0.0095
Credit_access	-0.150782	0.2405	-0.6300	0.5310	-0.6221	0.3205
Access_to_climate_inform	-0.44108**	0.5920	-0.7500	0.0060	-1.6014	0.7192
Extension_services	-0.018090	0.2964	-0.0600	0.9510	-0.5989	0.5628
_cons	-0.416121	1.0016	-0.4200	0.6780	-2.3792	1.5470

\*\*\*<1%, \*\*<5% and \*<10%

### Factors affecting smallholder farmers' crop productivity

The study carried out Cobb Douglas production analysis to determine factors affecting the productivity of crops (Table 9). Study results showed that income has a positive significant impact on crop productivity, productivity improves by 0.002%, with the outcome of increase in income level, which statistically significant at 0.0040 p-value ( $p < 0.01$ ). Fertilizer was found to have a significant positive impact on crop productivity. A unit increase in fertilizer use was associated with a 0.12% increase in crop yield, statistically significant at 0.0000 p-value ( $p < 0.001$ ). Farm size was found to harm crop productivity, the bigger the farm size, the lower the crop productivity by 6.52%. Livestock quantity was found to have a positive significant influence on crop productivity, the higher the number of livestock a farmer has, the higher the yield of crops



by 0.86%, statistically significant at 0.0001 p-value ( $p < 0.001$ ). Adopting CSA practices was found to have a profoundly favourable effect on crop productivity, if one more farmer adopts CSA practice, the average yield for the farmers improves by 13.49%, statistically significant at 0.0720 p values ( $p < 0.1$ ). The other factors were found not to have a significant impact on crop yield. Marital status influenced crop productivity by 0.07% while the education level of the household head had a 0.098% influence on contribution to crop productivity among smallholder farmers. Household size also contributed 0.2% to smallholder farmer crop productivity.

**Table 9.** Factors affecting smallholder farmers' crop productivity.

Linear regression		Number of Obs = 194				
					F(9, 179)	= 11.05
					Prob > F	= 0.0000
					R-squared	= 0.6441
					Root MSE	= 0.74495
log_yield	Coef.	Robust Std. Err.	t	P>t	[95% Conf. Interval]	
Age	-0.00192	0.0051	-0.3700	0.7090	-0.0120	0.0082
Gender	0.03854	0.1126	0.3400	0.7320	-0.1830	0.2601
Marital_status	0.00755*	0.0379	0.8410	0.0220	-0.0821	0.0670
Education	0.00980*	0.0122	0.8000	0.0420	-0.0338	0.0142
Farming_experience	0.00434	0.0049	0.8800	0.3770	-0.0053	0.0140
Household_size	0.02308**	0.0181	1.2800	0.0012	-0.0586	0.0124
Income	0.00002**	0.0000	2.9400	0.0040	0.0000	0.0000
Fertilizer	0.00123***	0.0002	8.1300	0.0000	0.0009	0.0015
Farm_size	-0.06518***	0.0145	-4.4900	0.0000	-0.0938	-0.0366
Livestockqt	0.00863***	0.0018	4.7900	0.0000	0.0051	0.0122
CSA_Practice	0.13490*	0.0747	1.8100	0.0720	-0.0120	0.2818
Credit_access	-0.11707	0.0741	-1.5800	0.1150	-0.2629	0.0287
Access_to_climate						
Inform	-0.15234	0.1974	-0.7700	0.4410	-0.5408	0.2361
Extension_services	0.04293	0.0846	0.5100	0.6120	-0.1236	0.2094
_cons	7.19355	0.3095	23.2400	0.0000	6.5845	7.8026

\*\*\*<1%, \*\*<5% and \*<10%

## DISCUSSION

The impact of CSA practice adoption on crop productivity among smallholder farmers and factors affecting adoption of smart climate-smart agricultural practices among smallholder farmers in Nyimba district were determined in this study. Among the factors affecting smallholder farmers' adoption of climate-smart agricultural practices, age, gender, farming experience, income, fertilizer use and livestock quantity were found to have a positive effect on CSA adoption while farm size and access to climate information had a negative influence on CSA adoption. The age category of 40-55 years and > 55 years to have adopted more CSAP in the study area. This indicates that most participants have long years of experience in the area which is helpful for farmers in climate change adaptation options including CSA. A study by Saha et al. (2019) and Zakaria et al. (2020) the level of agricultural experience is one of the factors for farmers choice of adaptation techniques for climate. Kurgat et al. (2020) showed that female ownership of farm assets, farm location, and household resources were major determinants of climate-smart agricultural adoption in Tanzania. A study by Aryal et al. (2018) concluded that factors such as household characteristics, market access, and main climate hazards are found to affect the probability and level of implementing different climate-smart practices of climate-smart agricultural adoption by smallholder farmers.

Concerning the factors affecting smallholder farmers' crop productivity, the results in this study showed that income, fertilizer and livestock quality are among the factors that have a positive significant impact on crop productivity. Livestock provides farming households with manure and animal draught power to produce crops and the investment of income from livestock into technologies that benefit crop production. In addition to the effects of manure and draught on crop output; money from livestock is frequently invested in terms that improve crop production. Mujeyi et al. (2021) found similar results on the adoption of climate-smart agriculture to significantly contribute to the crop yield of smallholder farmers in an integrated crop-livestock system. Marital status, education level of the household head and household size contribute to

crop productivity by 0.07%, 0.098% and 0.2% respectively. In a study by Serote et al. (2021) household demographics characteristics influenced the adoption of climate-smart agriculture and crop productivity.

Smallholder farmers' crop yields of CSAP adopters were 20.20% higher than for non-adopters. This study revealed that implementing CSAP increases maize yield for smallholder farmer adopters by 21.50% higher than non-adopters. Furthermore, including climate-smart agriculture practices in smallholder farmers' farming systems initiatives is critical for establishing resilient and sustainable farming communities. Prior research findings support our results; CSA practices can help resource-poor farmers become more resilient to climate change by increasing crop yields. A study by Abegunde et al. (2022) on the effect of climate-smart agriculture on household food security, also revealed that CSA practice adoption significantly and favorably affects household food security. The findings also indicated that agricultural revenue and income from non-farm sources had a significant impact on household food security (Abegunde et al., 2022).

Another study on the impact of climate-smart agriculture technology on productivity in southern Ethiopia showed that implementing CSA practice (row planting), had a significant impact on wheat yield among smallholder farmers' adopters (Mossie, 2022). Tadesse et al. (2021) conducted a study on the impact of climate-smart agriculture on soil carbon, crop productivity, and fertility in Ethiopia and revealed that climate-smart agriculture experimental fields showed yields 30–45% higher under CSA practice than the control ( $p < 0.05$ ).

Kichamu-Wachira et al. (2021), revealed similar results on the effect of CSAP to significantly increase crop yields among smallholder farmers in Africa. The study further concluded that CSAPs are an alternative advanced agricultural technology compared to conventional farming typologies due to their enhancement of food production through climate mitigation and improving soil quality. Furthermore, Amadu et al. (2020) found that 53% of CSAP adopters had increased yields of maize in the drought year of 2016 in southern Malawi. Fentie and Beyene's (2019) research findings from the PSM model revealed that the adoption of CSAPs had a significant impact on crop yield per hectare. Therefore, scaling up CSA will significantly

contribute to farmers' resilience to the adverse effects of climate change and climate variation by enhancing crop productivity and contributing to food security among farming households. Beedy et al. (2010) showed the significant and positive influence of *Gliricidia sepium* alley cropping on soil organic matter influence on the compiled single field of maize. Alley cropping had impacts on soil physicochemical properties in turn enhanced maize yields and increased soil nutrients over the mid-and-long term.

### CONCLUSION

The findings of this study support previous empirical studies' notion that the implementation of climate-smart agriculture improves crop productivity among adopter farmers. However, the adoption of CSA practices, despite its benefits, is not automatic among smallholder farmers, hence evaluating factors influencing CSA adoption and crop productivity in smallholder farming typologies is also important. This study found influencing factors such as farmer's level of education, household size, synthetic fertilizer usage, age of household head, gender, farming experience, livestock ownership, annual income, farm size, marital status of household head, and access to climate information, are significant determinants of CSA practice adoption and crop productivity.

The results of this study are crucial to the governmental and non-governmental organizations in Zambia especially those housed in Nyimba district with an interest in agriculture and working with smallholder farmers. This study provides a direction for policymakers to strengthen farmers' ability to climate-smart agricultural practices adoption through information sharing and policy reform around the agricultural sphere.

It is recommended that scholars undertake further similar research on the factors influencing CSA adoption and crop productivity in other parts of the country, but with more detailed inquiry, incorporating other indicators or variables not considered in this study and with a more holistic approach focussing on an independent CSA practice for a given farming typology.

### ACKNOWLEDGMENTS

This study was approved by the University of Zambia Directorate of Research and Graduate Studies (NASREC), and bears a NASREC IRB No. 00005465 (IORG No. 0005376). The authors thank the African Centre of Excellence for Climate Smart Agriculture and Biodiversity Conservation Management for their funding support through the World Bank, which enabled them to pursue the study.

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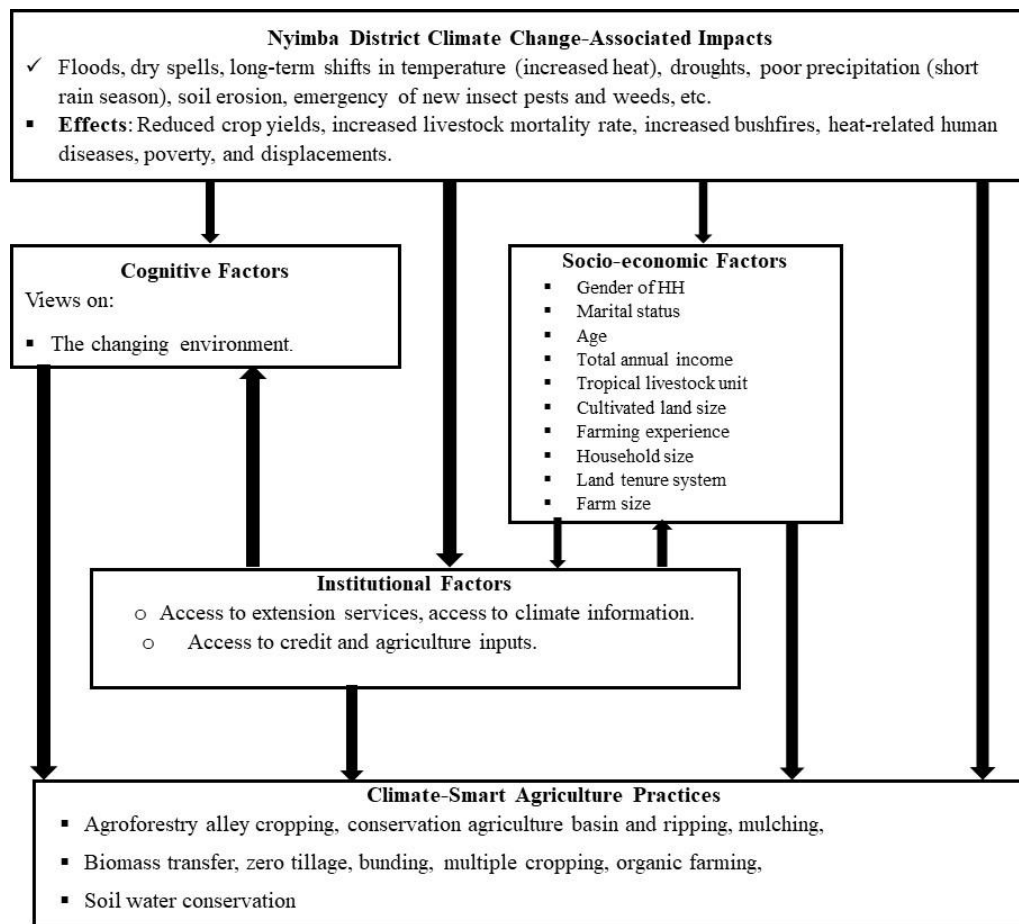
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**Annex 1.** Conceptual framework based on adoption.



Source: Adopted and modified from Serrat (2008).

# Ethiopian Journal of Biodiversity

## Guideline for contributors

### 1. Types of papers

- **Research papers** - Research papers should not exceed 8000 words in length, including Figures, Tables and References. Moreover, they should not contain more than 10 Figures and/or Tables
- **Review papers** - Critical and comprehensive reviews that provide new insights into or interpretations of the subject through a thorough and systematic evaluation of available evidence that should not exceed 10,000 words including Figures, Tables and References
- **Short communications** - Short communications such as opinions and commentaries should not exceed 1500 words and they must be brief definitive reports which need not be divided into Materials and Methods, Results and Discussions
- **Book Reviews** - Book review which is a critical evaluation of published books in any discipline of biological sciences/biodiversity will be published under this column

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#### 2.1 Article style and structure

Manuscripts should be written in American English, typed double-spaced, on A4 size, with margins of 1.5 cm on top and bottom sides of the paper, 2 cm on left and 1.5 cm on the right. A font size of 12 points (Times New Roman) should be used throughout the manuscript. The major sections of the manuscript include title, abstract, keywords, introduction, materials and methods, results, discussion, conclusion and recommendation, acknowledgements and references. Those sections having headings and sub-headings should not have more than three levels. All pages and lines should be numbered with the title page being page 1

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Desikan, S. and Ramesh, G. 2006. Software testing. Bangalore, India: Dorling Kindersley.

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Lemessa, D., Hylander, K. and Hambäck, P. 2013. Composition of crops and land use types in relation to crop raiding pattern at different distances from forests. *Agriculture Ecosystems and Environment*, **167:71-78.**

Mewded, B., Negash, M. and Awas, T. 2020. Woody species composition, structure and environmental determinants in a moist evergreen Afromontane forest, southern Ethiopia. *Journal of Forestry Research*, **31(4): 1173-1186**.

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Structure:

Last name, First initial. Year published. Article Title. *Journal*, [online] **Volume (Issue number): page(s)**. Available at: URL [Accessed Day Mo. Year].

Example:

Raina, S. 2015. Establishing Correlation Between Genetics and Nonresponse. *Journal of Postgraduate Medicine*, [online] **61(2):148**. Available at: <http://www.proquest.com/products-services/ProQuest-Research-Library.html> [Accessed 8 Apr. 2015].

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When no author is listed, use the following structure:

The website name, Year published. *Page title*. [online] Available at: URL [Accessed Day Mo. Year].

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**One author:** The last name of the author followed by year of publication will be cited in the text.

Example: .....(Brown, 2005).

**Two authors:** The last name of the authors are joined by "and" followed by year of publication.

Example: ..... (Tesfaye and Girma, 2019).

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Example :.....(Adugna et al., 2019).

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Tables should be as editable text and be placed on a separate pages at the end of the manuscript. Number tables consecutively (i.e. Table 1, Table 2, etc.) in accordance with their appearance in the text and avoid vertical lines and shading in the table cells. Table captions should be descriptive and appear above the table. Footnotes and sources to tables should be placed under the table. Larger datasets can be uploaded separately as Supplementary Files

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- A reference section containing all sources cited in the text
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**VOLUME 4, NO. 1 (APRIL 2023)**

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