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Assessment of the phenotypic purity and disease prevalence of improved barley (*Hordeum vulgare* L.) seeds collected from different sources from South and Central Ethiopia

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Abstract: **Barley is one of the major cereal crops grown in the high lands of Ethiopia that serves as sources of food and income. Despite its usage, the national average yield is still very low as compared to its potential due to lack of quality seed that is mainly caused by losses of phenotypic purity are the major biotic constraints on barley production. Thus, the aim of this study was to assess the phenotypic purity and diseases prevalence of improved barley seeds collected from different sources in South and central Ethiopia. The assessment was done at six zones and two agricultural research centers of major barley growing areas of South and Central Ethiopia. The field experiment was conducted at Werabe agricultural research center experimental station in a Randomized Complete Block Design with three replications. The treatments were arranged based on varieties and its seed classes. True to type and seed testing standard was used as standard check. The study found that the majority of farmers used HB1307 (70.4%), followed by Ibon174/03 (16%) and HB42 (13.6%). Grow-out tests revealed that both collected samples and standard checks lacked phenotypic purity, containing mixtures of different cultivars. Off-type rates in nucleus seeds were 2.4% for HB1307, 3% for HB42, and 2% for Ibon174/03, with additional contamination from other varieties. Higher seed classes, such as nucleus and breeder seeds, demonstrated better purity and lower disease incidence than standard seed classes. The findings highlight the need for stricter national seed quality control and suggest further multi-seasonal studies to validate results and guide policy improvements.**

Keywords: **Barley seed classes, grow out test, phenotypic purity.**

1. INTRODUCTION

Ethiopia is mainly an agrarian country with the vast majority of population directly or indirectly involved in crop production (Tadesse *et al*., 2021). Agriculture plays a significant role in the life and livelihood of most Ethiopians, about 12 million household farming's account approximately 95% of crop production (FAO, 2011). It contributed 34.1% to the GDP, creates employment opportunities for 79% of the population, responsible for 79% of external earnings and major sources of row material and wealth for the investment and market (Diriba, 2020). Despite its role, there has been low due to low level capacity use of improved technologies by the majority of the farmers (Abebe *et al*., 2017; CSA, 2013; Tarekegn and Mogiso, 2020). Owing to that, food security issues facing to Ethiopia year to year sense of urgency underlying the need for enhancing both seeds and yields (Tarekegn and Mogiso, 2020).

Barley (*Hordeum vulgare* L.), is one of the most important and top ten cereal crops in the world after wheat, maize and rice (Samuel, 2016). It grows in diverse agro ecologies from 1800 to 3400 meter above sea level, produced at approximately

4.1million smallholder farmers and it serves as sources of food and income (Muluken, 2013; Shahidur *et al* 2015; Ganewo *et al*., 2022; Alemayehu, 2024). It is also makes Ethiopia being the 2nd largest producer in Africa next to Morocco, accounting 25% of the total barley production in the world (FAO, 2014). In the year of 2020/2021, Ethiopia's, barley production was estimated 2.34 million tons, cultivated at approximately 0.92 million hectares of land (CSA, 2021). Despite its usage and large area under production, the national average yield is still very low 2.11 to 2.53 tons ha^{-1} as compared to world 2.89 tons ha^{-1} and its potential 6 tons ha^{-1}(CSA, 2021, 2023).

In Ethiopia, lack of quality seed production and associated technologies could be mentioned among the major challenges that hindered barley production and productivity (Abebe *et al*., 2017; Bekele *et al*., 2019). Seed is the most important agricultural input for improved varieties and widely recognized as the fundamental to ensure increment of crop production and productivity (Kassa and Merkine, 2020) and it is also the basic unit for distribution and maintenance of plant population (Gebremedhin, 2015; Kassa and Merkine, 2020; Golijan *et al*., 2024), in terms of agriculture it is the renewal of plant production (Popovic, 2010). To enhance agricultural productivity, food security and reduce poverty, quality seed is highly acknowledged (Golijan *et al*., 2024). Generally, for successful crop production, the use of good quality seed can increase the yields from 15 to 20% (Ambika *et al*., 2014). That's why ensuring high seed quality is priority of current seed production and a prerequisite for high yields of all plant species (Postic *et al*., 2014). Inherent, loss of phenotypic purity are the major biotic constraints on barley production in Ethiopia (Wondimu *et al*., 2022). Therefore, high level of phenotypic purity and disease free crop varieties must be achieved and maintained for the improvements of productivity and seed quality imparted by breeders. Thus, the aim of this study was to asses the phenotypic purity and disease prevalence of improved barley seeds collected from different sources and to determine the phonological purity status and extent to which the submitted sample confirms to the prescribed standard.

2. MATERIALS AND METHODS

The assessment and evaluation using grow out test was done in major barley growing areas of the previous South Ethiopia and Werabe agricultural research center's experimental station during 2022 main cropping season. Six Zones and two districts from each four zone (Siltie, Gurage, Gamo, Hadya) and one district from Kembata Zone); total 9 districts (eight farmers from each), and two research centers, Werabe (two samples) and Holeta agricultural research centers (six samples in addition to their true to type) a total of 81 sample were collected and assessed in this study. Primary data were collected from the respondents of the selected surveyed areas. Different samples of improved barley seed varieties were collected from interviewed farmers, unions and research centers based on the trend of improved seed usage, seed classes and their standard checks which was intended for the grow out test in the field experiment. Seed samples collected from the selected surveyed areas were bulked according to the identified varieties and its seed classes and subjected to field experiment.

The treatments were arranged based on the collected barley varieties and seed classes (food barley HB1307 (5 seed classes), HB42 (3 seed classes) and malt barley Ibon174/03 (6 seed classes) were laid out in the Randomized Complete Block Design (RCBD) with 3 replications. Nucleus seed of the tested varieties and seed testing standard (ISTA Rule) (Barsa, 2002) was used as standard check. The recommended row length of 6m, spacing between row 0.25m and space between plots 0.50 m in a plot size of 6 m x 1.2m (total area of 7.2 m2) were used by hand drill in the row. Planting time was launched at the first of July based on crop calendar of the areas. The trial was fertilized with recommended rate of 100 kg/ha NPS during planting and 100 kg/ha Urea with two time split application.

To assess conventional phenotypic purity, observations were made on 400 plants per treatment, focusing on the presence of off-types, other crop species, or different barley varieties. Key morphological traits such as row type, leaf auricle, collar region color, internode color, ear head structure, plant height, flag leaf angle, ligule size, stigma color and maturity were closely monitored. Additionally, disease symptoms including head smut, leaf rust, scald, spot blotch, and net blotch were recorded. The grow-out test plots were evaluated throughout the entire growing season, with particular attention given to the flowering and maturation stages.

Each plant was scrutinized for distinguishing features characteristic of the barley cultivar under both test and control conditions (true-to-type). Any plant exhibiting deviations in the specified traits was tagged for further inspection. These anomalies were later confirmed as either off-types or other crop species, documented accordingly, and subsequently removed from the plots.

Data collected through field experiments and questionnaires were analyzed using SPSS software. Phenotypic purity was calculated and interpreted based on the International Seed Testing Association (ISTA, 2004) standards. The percentage of phenotypic purity was calculated by comparing the number of pure plants to the total number of sampled plants grown. Off-type plants (those that didn't match the variety's description) were identified and counted to determine the phonological purity of the seed lot. The percentage of off-type plants was calculated to determine the phenotypic purity of the seed lot.

Table 1. The minimum standards for phenotypic purity detection for different seed classes used in this study

No	Classes of seeds	Purity %
1	Breeder seed	100
2	Foundation seed	99
3	Certified seed	98
4	Hybrid seed	95

Source: (Basra, 2002)

Table 2. The minimum certification standard for phenotypic purity and maximum permissibility of off-types on the number of plants required per sample for barley crop

No	Maximum permissible off-types %	Minimum phenotypic purity%	No of plants required/sample for observation
1	0.10	99.9	4000
2	0.20	99.8	2000
3	0.30	99.7	1350
4	0.50	99.5	800
5	**1.0 and above**	**99.0 and below**	**400***

Source: (ESE, 2025)

3. RESULTS AND DISCUSSIONS

Response of farmers based on barley varieties grown, seed classes and storage pests in the surveyed areas

The results of the study revealed that during the assessment, most of the respondents produce food barley HB1307 (70.4%), HB42 (13.6%) and malt barley Ibon174/03 (16%). Use of improved barley seeds in the assessed areas was also limited due to shortage of quality seeds, except the research centres. This result is in agreement with the finding of (Samuel, 2016) that reported, Ethiopian farmers grown two types of barley food and malt barley, majority of barley that farmers grow was food type, which was in lined with finding of (UK, 2018) who reported that in Ethiopia improved varieties of certified seed of barley were not always available. The response of farmers regarding to storage pests, 32.1% (26) and 4.9% (4) of the respondent's stored barley product was damaged by rodents and weevil, respectively (Table 3).

Table 3. The response of farmers based on the variety produced, seed classes and storage methods

Barley varieties grown in the surveyed areas	Frequency	Percent
Food barley (HB1307)	57	70.4
Food barley (HB42)	11	13.6
Malt barley (Ibon 174/03)	13	16.0
Seed class		
Nucleus seed (HB1307, HB42, Ibon 174/03)	3	3.7
Breeder seed (HB1307, HB42, Ibon 174/03)	3	3.7
Pre basic seed (HB1307, Ibon 174/03)	2	2.5
Basic seed (Ibon174/03)	1	1.2
C1(Certified seed one) (HB1307, Ibon174/03)	40	49.4

C2 (Certified seed two) (HB1307, Ibon174/03)	23	28.4
C4 (Certified seed four) (HB42)	9	11.1
Storage pest		
Rodents	26	32.1
Rodent and weevil	4	4.9
No	51	63.0
Total sample size	81	

Source: (Own survey data, 2023)

Reaction of farmers regarding to storage methods used

The findings of this study revealed that, with the exception of unions and research centers, most of the surveyed farmers stored their harvests in traditional granaries (Fig 1). This practice emerged as a key contributor to seed deterioration, primarily due to elevated moisture levels. Research from Ethiopia and other sub-Saharan African regions consistently highlights a strong dependence on conventional storage methods. Ariong *et al.* (2023) reported that poor harvest and postharvest handling led to approximately 22% loss in both yield and quality. These traditional techniques often fail to maintain the optimal grain moisture threshold of ≤13%, which critically affects storage longevity. Similarly, Abbas *et al.* (2014) emphasized that high temperatures and excessive seed moisture linked to relative humidity are major drivers of seed degradation and reduced viability in such storage systems.

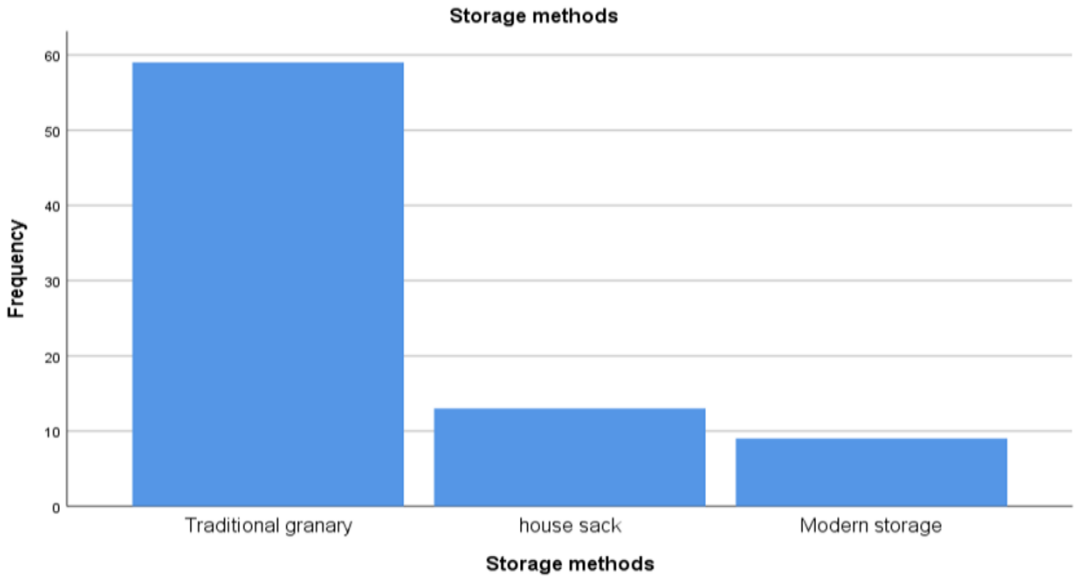

Figure 1. Response of farmers to storage methods used

The phenotypic purity of improved barley seed samples collected from different sources under field experiment using grow-out test

Grow-out test results revealed that both the collected barley samples and their standard checks were phenotypically mixed, indicating the presence of multiple cultivars rather than a single pure variety. This lack of uniformity compromises the reliability of genetic analyses and varietal comparisons. According to ISTA (2004) standards, the phenotypic purity of nucleus seeds was recorded as 99.4% for HB1307, 97.05% for HB42, and 99.65% for Ibon174/03, with corresponding off-type rates of 2.4%, 3%, and 1.4%. Additional observations showed the presence of other cultivars at rates of 1.8%, 11.4%, and 1.4%, respectively, further confirming that even control samples lacked full purity (Table 4). These findings align with previous studies (Bekele et al., 2019; Garvin & Carver, 2003), which reported variability in seed quality across different sources and classes, and highlighted genetic contamination as a challenge in crop improvement. Moreover, the Ethiopian seed certification standards (QSAE, 2012) set strict limits for seed-borne disease infections ranging from 0% for breeder

and pre-basic seed, 0.05% for basic seed, 0.2% for certified (C1 to C4) and 0.4% for emergency seed. Different seed sources and classes varied on physical and physiological seed quality parameters. But in this study all samples collected and their control were above the threshold limit as compared to the national seed healthy standard permitted in certified seeds. This might be due to the association of the fungi with seeds from the sources, sub optimal storage conditions and favourable climatic conditions for the development of pathogens in the field during growth of the crop.

Table 4. Interpretations of the results of grow out test using standard levels

No	Treatments	MNOT%	MNOC%	Scald	SB	NB	HS	RI response	PhP%	Results
1	Nu HB1307	2.4	1.8	4	12.5	3	0	2.5TR	99.55	NP
2	BR HB1307	11.4	2.4	4	17.5	3	0	2.5TR	99.4	NP
3	PB HB1307	12.8	2.1	4	17.5	4	0	5R	97.9	NP
4	C1 HB1307	17.8	2.1	3	17.5	4	3	5R	97.9	NP
5	C2 HB1307	25.4	2.85	3	17.5	5	5	5R	97.15	NP
6	Nu HB42	3	2.95	5	12.5	3	0	2.5TR	97.05	NP
7	BR HB42	14	2.5	5	17.5	5	0	5R	97.5	NP
8	C5 HB42	27	27.35	10	20	10	8	12.5MRMS	72.65	NP
9	Nu Ibon174/03	2	0.35	5	11.5	3.5	0	2.5TR	99.65	NP
10	BR Ibon174/03	4.4	2.75	7.5	16.5	5	0	5R	97.25	NP
11	PB IBON	12, 3	4.35	7.5	22.5	6	0	7.5MR	95.65	NP
12	B Ibon174/03	10. 8	6.6	7.5	22.5	7.5	0	7.5MR	93.4	NP
13	C1 Ibon174/03	18	15.75	10	27.5	10	1	20MRMS	84.25	NP
14	C2 Ibon174/03	21	24	12.5	27.5	12.5	1	27.5MRMS	76	NP

NB: MNOT=Mean No off- types, MNOC=Mean No of other cultivars, SB= Spot. Bloch, NB= Net Bloch, HS= Head smut, Nu = nucleus, B= breeder, PB= pre basic, B= basic C= certified seed, PhP%= Phenotypic purity percentage, NP= not pure, MR= moderate resistance, MRMS =moderate resistance to moderate susceptible, R=resistance, TR =Thrace, RI= rust infection

4. CONCLUSION AND RECOMMENDATIONS

Most of barley producer farmers of the surveyed areas used their own saved seeds of improved barley varieties (HB1307, Ibon174/03) for many years which had its own impact for the reduction of yield and quality of the production due to mainly physical contaminations. Grow-out test evaluations based on ISTA standards, both the collected samples and the standard check (true to type) exhibited phenotypic impurity, indicating a mixture of different cultivars rather than uniform varietal identity. The authenticity of the cultivars, as determined by phenotypic traits, declined with lower seed classes when compared to the control and standard benchmarks. In contrast, seeds sourced from higher-level classes such as nucleus and breeder seeds demonstrated superior phenotypic purity and lower incidence of disease, highlighting their reliability over standard seed classes.

This result showed that the phenotypic purity of the varieties reduced as the seed classes lower, due to physical deterioration and susceptibility to disease.Therefore, this assessment would strongly be commended to str engthened seed phenotypic purity and diseases aspects in order to increase the productivity of barley crop. Prevent pathogenic seed borne diseases transmitted from seed to seedling of the next generation. Thus, the national seed policy should insure the collected plant phenotype and evaluated by the national research and development program. Since the findings were constrained to a single season multi-seasonal investigation might be suggested.

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Author contributions

AF, MM, MK and DA designed the research; all authors conducted the experiment and collected the data; AF analyzed, interpreted the data and wrote the full manuscript. All authors read, reviewed and approved the final manuscript.

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Declarations

Competing interests: The authors declare that they have no competing interests.

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