

Effect of storage condition and storage duration on grain quality of wheat

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Summary

The storage conditions and duration of harvested grain storage may have a significant impact on the concentration of essential nutrients like iron, zinc, and protein in wheat. Poor storage practices, especially in environments with high humidity and temperature, can lead to nutrient degradation and loss of grain quality over time. The objective of this study was to understand if the storage duration and conditions of the storage affects nutritional concentration (zinc, iron and protein) in wheat compared to sample analysed shortly after harvest. The study was implemented for wheat grain in ICARDA quality laboratory, Rabat, Morocco in 2023/24. This study considered two major factors (i) grain storage duration (6 treatments, i.e., immediately after harvest, and an interval of 30, 60, 120, 180, and 360 days after harvest) and (ii) three different storage environments, i.e., room temperature, stored in refrigerator (4°C), and in Freezer (-20°C). The study found that there was no significant effect of storage duration and storage temperature on the concentration of iron, zinc in bread wheat grains. This suggests that if samples may not be possible to process for analyses immediately after harvesting it can be safely stored and analyzed in the convenient time within a year (as the study covers up to 360 days) without affecting these nutrient levels. However, grain protein content was slight reduction after 12 months of storage, i.e., grain protein was reduced by 0.87 (by 5.92%) when stored at room temperature, by 0.97 (6.6%) when stored at 4°C (at refrigerator and reduce by 1.07 (7.28%) when stored at freezer (-20°C) compared to the grain protein obtained immediately after harvest (14.7%).

1. Introduction

Over two billion of the world's population is at the risk of nutrient deficiency which is linked to inadequate supply of nutrient in daily diets (Cakmak & Kutman, 2018; Passarelli, *et al.*, 2024). This is because the major foods consumed have low concentrations of essential nutrients, associated with poor soil quality and/or crop management practices, and crop and variety used. Still, most crop management efforts focus on increasing productivity and not so much on issues related to malnutrition. Also, agronomy R&D typically focuses on social, economic, and environmental outcome indicators. Nutritional outcome indicators (e.g., the nutrient content in the harvested product) which are important for improving nutritional security are often not considered. Recent research points to the potential for agronomic interventions to improve the nutritional quality of produce but guidelines on the assessment

can be made have been missing. Besides genetic and crop management factor methods of nutrient analysis, the way sample process for nutrient analysis in the laboratory may also affect nutrient concentration in the harvested products. Maintaining the nutritional condition of the stored grain is important for major staple crops. Storage conditions and the duration of grain storage may have a significant impact on the concentration of essential nutrients like iron, zinc, and protein in wheat. Poor storage practices, especially in environments with high humidity and temperature, can lead to nutrient degradation and loss of grain quality over time (Ziegler *et al.* 2021). Prolonged exposure to such conditions can trigger biochemical changes, oxidation, and enzymatic activities that reduce the bioavailability of micronutrients like iron and zinc. Protein content and quality may also decline, as proteins are susceptible to denaturation and breakdown when exposed to adverse storage conditions. Moreover, improper storage can lead to infestation by pests and mold growth, which further compromise the nutrient profile and safety of the grain. By maintaining optimal storage conditions, the concentrations of iron, zinc, and protein are better preserved, ensuring that wheat remains a valuable source of these essential nutrients for human consumption, especially in regions where wheat is a primary dietary staple.

Logistically, it is not always possible to analyse samples immediately/in a few days after the crop harvest, however it is not known if the storage condition and its duration affect grain nutrient concentration (Fe, Zn and protein) compared to sample analysed few days after crop harvest. Hence, the objective of this experimentation is to understand if the storage duration and condition of storage affects nutritional concentration (zinc, iron and protein) compared to sample analysed in a few days of crop harvest, specially.

2. Research Methodology

2.1 Study location

The study was conducted in International Center for Agriculture Research for Dry Areas (ICARDA)'s quality assessment laboratory in Rabat, Morocco during 2023/24. Bread wheat (*Triticum aestivum L.*) was selected as a grain crop to evaluate for this study.

2.2 Collection of grain samples

For this study, homogeneous wheat grain sample was taken from the 2022/23 wheat growing season from a grain production field of ICARDA research station Merchouch. A grain sample of 10 kg was taken immediately after harvest. The grain sample was sun-dried and well cleaned by removing off-

type, chaffs and other inert material. From this homogenous sample subsample of 150 g was prepared for each treatment. Samples were well packed in paper bags and were kept in zip-lock bags.

2.3 Experimental treatments and design

To understand storage duration and storage condition on grain quality (iron, zinc and protein concentration), the following treatments were examined.

Treatment details

Factor 1: Storage duration: following six storage duration was included for this study:

- i. Immediately after harvest,
- ii. 30 days after harvest
- iii. 60 days after harvest
- iv. 120 days after harvest
- v. 180 days after harvest
- vi. 360 days after harvest

Factor 2: Storage condition: Following three storage environments were included in this study.

- i. At Room temperature
- ii. At Refrigerator (4°C)
- iii. At Freezer (-20 °C)

Total number of treatment: 19 (six storage duration × three storage environment and including a control treatment, i.e., analysis immediately/few days after harvest).

Treatment combinations:

1. 30 days after harvest: at Room temperature
2. 60 days after harvest: at Room temperature
3. 90 days after harvest: at Room temperature
4. 120 days after harvest: at Room temperature
5. 180 days after harvest: at Room temperature
6. 360 days after harvest: at Room temperature
7. 30 days after harvest: at Refrigerator (4°C)
8. 60 days after harvest: at Refrigerator (4°C)
9. 90 days after harvest: at Refrigerator (4°C)
10. 120 days after harvest: at Refrigerator (4°C)
11. 180 days after harvest: at Refrigerator (4°C)
12. 360 days after harvest: at Refrigerator (4°C)
13. 30 days after harvest: at Freezer (-20 °C)
14. 60 days after harvest: at Freezer (-20 °C)

15. 90 days after harvest: at Freezer (-20 °C)
16. 120 days after harvest: at Freezer (-20 °C)
17. 180 days after harvest: at Freezer (-20 °C)
18. 360 days after harvest: at Freezer (-20 °C)
19. Control: grain analysis within few days after harvest.

Number of replications: 3

Experimental Design: Completely Randomized Design (CRD)

Table 1 Summarizes the treatment detail including storage conditions and storage duration of the samples before analysis.

Sample	Rep	Immediately after harvest (Control)	Storage duration	Storage conditions			
				Room temperature (F1)	Refrigerator (4°C) (F2)	Freezer (-20°C) (F3)	
Bread wheat (BW)	R1	Control R1	T1	30 Days after harvest	F1T1R1	F2T1R1	F3T1R1
			T2	60 Days after harvest	F1T2R1	F2T2R1	F3T2R1
			T3	90 Days after harvest	F1T3R1	F2T3R1	F3T3R1
			T4	120 Days after harvest	F1T4R1	F2T4R1	F3T4R1
			T5	180 Days after harvest	F1T5R1	F2T5R1	F3T5R1
			T6	360 Days after harvest	F1T6R1	F2T6R1	F3T6R1
	R2	Control R2	T1	30 Days after harvest	F1T1R2	F2T1R2	F3T1R2
			T2	60 Days after harvest	F1T2R2	F2T2R2	F3T2R2
			T3	90 Days after harvest	F1T3R2	F2T3R2	F3T3R2
			T4	120 Days after harvest	F1T4R2	F2T4R2	F3T4R2
			T5	180 Days after harvest	F1T5R2	F2T5R2	F3T5R2
			T6	360 Days after harvest	F1T6R2	F2T6R2	F3T6R2
	R3	Control R3	T1	30 Days after harvest	F1T1R3	F2T1R3	F3T1R3
			T2	60 Days after harvest	F1T2R3	F2T2R3	F3T2R3
			T3	90 Days after harvest	F1T3R3	F2T3R3	F3T3R3
			T4	120 Days after harvest	F1T4R3	F2T4R3	F3T4R3
			T5	180 Days after harvest	F1T5R3	F2T5R3	F3T5R3
			T6	360 Days after harvest	F1T6R3	F2T6R3	F3T6R3

2.4 Laboratory analysis

Grain samples were analyzed in ICARDA's grain quality assessment laboratory which is located in Rabat, Morocco.

Iron and Zinc concentration: For measuring grain Iron and Zinc concentration, we have used non-destructive X-ray Fluorescence (XRF) spectroscopy as a non-destructive analytical method. The XRF device was well calibrated for bread wheat iron and zinc content before the analysis.

Calibration of non-destructive X-ray Fluorescence (XRF) spectroscopy for iron and zinc concentration: First, a representative set of samples is analyzed using ICP-OES (Inductively Coupled Plasma Optical Emission Spectrometry) to obtain precise lab-based measurements of the elements of

interest, such as iron and zinc. These same samples are then scanned using the XRF instrument. The final step is to correlate the element concentrations predicted by the XRF scans with the actual values obtained from the ICP-OES measurements. This correlation forms the basis for a calibration model.

Measurement of Protein content: The Kjeldahl method was used to determine total nitrogen content, and thus crude protein levels, by converting nitrogen into a quantifiable form through digestion, distillation, and titration. We have used method used by Baethgen and Alley et al. (1989) for measuring nitrogen content and converted percent protein content as shown in equation (1).

The protein concentration is determined by the following formula:

$$\% \text{ Protein: } (D.O * 12.5 * 14) / (0.024 * \text{weight} * 18) * 5.8 \dots\dots\dots \text{Equation (1)}$$

where,

- **D.O:** Optical Density at 650 nm
- **12.5 mL:** Final volume during colorimetric assay
- **14 g/mol:** Molar mass of nitrogen
- **0.024:** Slope of the standard curve
- **18 g/mol:** Molar mass of ammonium (NH₄⁺)
- **5.8:** Conversion factor from nitrogen content to protein content in bread wheat



Statistical analysis: We have used GenStat Twenty-third Edition for analysis of variance (ANOVA).

3 Results and Discussion

3.1 Effect of storage duration on grain Zinc (Zn), Iron (Fe) and protein concentration in wheat

In the sample we study, the average grain zinc (25.0 mg/kg) and iron (45.61 mg/kg) concentration at harvest are far below the nutritional target set by harvest plus (Zn: 38.5 mg/kg and Fe: 58.9 mg/kg) for wheat. The average protein concentration was 13.9% (Table 3). Irrespective of the storage temperature, there was no significant effect of storage duration in grain zinc and iron concentration for bread wheat compared to the control, i.e., analyzed samples in a few days after crop harvest (Table 2). Although there was a slight decreasing trend of Zn, Fe and protein concentration after 12 months of grain storage compared to control. This indicates that storage duration (up to 12 months) does not significantly impact the grain Fe, Zn and protein concentration in wheat grain in rainfed drylands (Table 3).

Table: 2. Analysis of variance (ANOVA) showing significant level (at $p=0.05$) of storage condition, storage duration and their interaction on grain iron, zinc and protein concentration in bread wheat.

Numbers in the table is significant level at $p=0.05$.

Source of variation	DF	Iron	Zinc	Protein
Replication	2			
Storage duration (SD)	5	0.857 (ns)	0.55 (ns)	<.001 (***)
Storage environment (SE)	2	0.766 (ns)	0.85 (ns)	<.001(***)
SD x SE	10	0.817 (ns)	0.12 (ns)	0.694 (ns)

Table 3. Effect of storage duration on grain zinc, iron, and protein concentration in wheat grain

Storage duration	Grain Zinc (mg/kg)	Grain Iron (mg/kg)	Protein content (%)
Immediate after harvest	25.64	46.16	14.7 ^a
30 days after harvest	24.18	45.40	13.90 ^b
60 days after harvest	25.60	44.88	13.94 ^b
90 days after harvest	24.94	46.01	13.81 ^{bc}
120 days after harvest	24.63	46.94	13.96 ^b
180 days after harvest	24.60	43.87	13.97 ^b
360 days after harvest	24.94	45.54	13.73 ^c
LSD	ns	ns	0.13

3.2 Effect of storage temperature on grain Zn, Fe and protein concentration in wheat

Irrespective of the storage duration, grain Zn and Fe concentration in wheat did not affect significantly due to storage temperature (zinc $p=0.7$; iron $p=0.6$), i.e., sample stored at room temperature, stored at

Refrigerator (4⁰C) and cold storage (-20⁰C) (Table 4). Although there is no significant difference, compared to store at room temperature, sample stored at cold storage has slightly higher Zn and Fe concentration. This indicates if samples could not be analyzed immediately after harvesting it can be stored either at refrigerator or at cold storage to maintain Zn and Fe concentration of grain samples to be analyzed for Fe and Zn.

There was a significant effect (p=0.01) of storage environment on grain percent protein concentration in wheat. Compared to the grain stored at room temperature (14.04%), grain protein content was reduced when grain sample stored at cold storage (-20⁰C) (13.85%) and at refrigerator (4⁰C) (13.91%) (Table 4). However, there was no difference in grain protein content between sample stored at refrigerator (4⁰C) and stored at cold storage (-20⁰C). This indicates that storage temperature affects grain protein concentration and has to take account if wheat grain samples could not be analyzed immediately after harvest.

Table 4. Effect of storage temperature on grain zinc, iron, and protein concentration in wheat grain

Storage condition	Grain Zinc (mg/kg)	Grain Iron (mg/kg)	Protein content (%)
Room temperature	24.69	45.35	14.04 ^a
In Refrigerator (4 ⁰ C)	24.93	45.30	13.91 ^b
In cold storage (-20 ⁰ C)	25.26	46.07	13.85 ^b
Control	25.64	46.16	14.7
LSD	ns	ns	0.09

Note: ns=non-significant; LSD= least significant difference

3.3 Interaction effect storage temperature and storage duration on grain Zn, Fe and protein concentration in wheat

There was no significant interaction effect between storage condition and storage duration on grain Fe and Zn concentration in bread wheat (Table 5). However, grain protein concentration was affected by interaction between storage temperature and duration of the storage. There was a slight reduction in grain protein concentration over the time in all storage conditions over the time period. After 12 months of storage, grain protein was reduced by 0.87 (by 5.92%) when stored at room temperature, by 0.97 (by 6.6%) when stored at 4⁰C (at refrigerator) and reduce by 1.07 (7.28%) when stored at freezer (-20⁰C) compared to the grain protein obtained immediately after harvest (14.7%) (Table 5).

Table: 5. Interaction effect of storage temperature and storage duration on grain zinc, iron and protein concentration in wheat grain.

	Grain Zn (mg/kg)	Grain Fe (mg/kg)	Grain protein (%)
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Storage duration	Room Temp.	Temp. at 4°C)	Temp. at -20°C)	Room Temp.	Temp. at 4°C)	Temp. at -20°C)	Room Temp.	Temp. at 4°C)	Temp. at -20°C)
After harvest	25.64			46.16			14.7		
30 DAH	23.43	22.87	26.25	45.31	43.13	47.67	14.0	13.9	13.80
60 DAH	24.37	26.81	25.63	45.27	45.28	43.98	14.3	14.2	14.10
90 DAH	25.85	24.53	24.42	46.24	45.46	46.25	13.87	13.83	13.73
120 DAH	23.65	25.16	25.09	45.03	46.05	49.63	14.07	13.97	13.87
180 DAH	23.30	24.89	25.62	45.42	44.96	44.12	14.16	13.80	13.97
360 DAH	24.55	25.33	24.54	45.83	45.93	44.78	13.83	13.73	13.63

4 Summary

The study highlighted that there is no significant effect of storage duration and storage temperature on bread wheat grain iron, zinc concentration. This suggest that if samples may not be possible to process for analyses immediately after harvesting it can be safely stored and analyse in the convenient time within a year (as the study cover up to 360 days). However, grain protein content was slightly reducing after 12 months of storage, i.e., grain protein was reduced by 0.87 (by 5.92%) when stored at room temperature, by 0.97 (6.6%) when stored at 4°C (at refrigerator and reduce by 1.07 (7.28%) when stored at freezer (-20°C) compared to the grain protein obtained immediately after harvest (14.7%).

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