Mineral Uptake, Total Polyphenols and Total Flavonoids of Barley as Affected by Tillage Practices under Semi-Arid Conditions

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Authors’ contributions

This work was carried out in collaboration among all authors. Authors NC, AC, ZBAI and MBH designed the study, performed the statistical analysis and wrote the protocol. Author NC wrote the manuscript. Authors AC and ZBAI installed the trials and applied necessary management practices. All authors read and approved the final manuscript.

ABSTRACT

Aims: Conservation agriculture has been recommended as an option to mitigate climate change impact when practicing conventional, to ensure sustainability and food security. This study examined the effect of conventional tillage (CT) and no tillage (NT) on mineral elements uptake, total phenolic content (TPC) and total flavonoid content (TFC) of barley.

Study Design: Split-plot design was applied for this study.

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Place and Duration of Study: The experiment was conducted in ESAK station (Boulifa, kef, North West Tunisia) during 2016/2017 cultivation year.

Methodology: Mineral elements uptake, total phenolic content (TPC) and total flavonoid content (TFC) of barley were studied as affected by conventional tillage (CT) and no tillage (NT) for tillering and grain filling stages.

Results: The results showed that tillage practices (T) had no significant effect on mineral uptake, total phenolic content and total flavonoids content under rainfed conditions. The stage (S) had showed significant effects on P, Ca and Na amounts for both tillage practices when it had no effect on K amount. The interaction T x S had no significant effect on mineral elements concentrations and TFC. However, this interaction had affected TPC significantly. Phosphorous (P) showed high significant positive correlations with Ca and Na. In addition, P presented high significant negative correlations with TPC and TFC. For partial correlation based on Tillage practices, similar correlations values were noted. Considering the partial correlation based on plant stages, no significant correlations had been noted.

Conclusion: This work enlarges our knowledge on barley mineral elements uptake, TPC and TFC as influenced by tillage practices aiding decision makers in increasing no tillage adoption in Tunisia under rainfed conditions.

Keywords: Tillage practices; plant stage; barley; mineral elements; TPC; TFC.

1. INTRODUCTION

World population increase and food global demand evolution requires a satisfying and sustainable crop production. In the Mediterranean area, one of the oldest and main cultivated crops is barley (*Hordeum vulgare* L.). It is one of the most cultivated cereal in Tunisia, Africa and World occupying respectively 544000 ha, 5514379 ha and producing 478000 tonnes, 6566569 tonnes and 148478878 tonnes, respectively [1]. In Tunisia, barley is essentially cultivated in regions with arid and semiarid climates that present annually less than 400 mm of rainfall [2]. As winter fast-growing cereal, barley could be cultivated as cover crop for soil preservation and as a forage plant [3]. Its resistance to drought and various environmental stresses result in its worldwide cultivation [4]. Only 2% of cultivated barley is directly consumed by humans, about 65% are used for animal feed and 33% for malting [5]. Barley regular consumption has been related to the risk reduction of certain diseases such as high blood pressure [6], heart problems [7], and colonic cancer [8]. Barley mineral elements, proteins and fiber component are mainly recognized for barley’s health advantages [9].

Besides, different classes of phytochemicals have been known in barley, and their activities have been noted [10]. Some of barley phytochemicals have presented health benefits such as flavonoids, phenolic acids, lignans, folates and sterols [11]. Phenolic compounds contribute to essential functions in plant reproduction and growth, and to plant defense system against insects, fungi and nematodes [12]. However, all along its growth, plant chemical and biochemical composition could fluctuate as affected by environments factors. Management practices such as tillage have been shown to influence nutrient content [13]. Cereal antioxidant contents could differ according to the environment, the genotype and probably genotype-environment interactions [14-15]. As well, climates changes added to the problems of cultivars adaptation to agro-ecological zones and unbalanced socio-economic conditions threaten barley production sustainability.

Commonly, farmers till their soils before sowing cereals in conventional agriculture to deal with weeds, to prepare seedbed and to avoid crust formation. Nevertheless, these practices added to climate change and monocultures lead to soil moisture loss and soil organic matter reduction [16] and erosion [17].

Consequently, conservation agriculture emerged as a substitution to conventional agriculture. Then, no tillage was adopted for the first time in 1999 in North West Tunisia under rainfed conditions [17]. About 260000 ha of agricultural area have been designed as a priority for conservation agriculture adoption in Tunisian semi-arid and sub-humid regions [18].

Since the worldwide adoption of conservation agriculture, research focused on its effect on soil organic matter, soil physical properties, soil moisture and yield [19]. This effect is related to weather conditions, rainfall, crop sequence, and the interaction between many other factors [20-
These interactions could result in stability, decrease or increase of grain yield [22]. As well, no tillage is recognized to ameliorate soil moisture, soil physico-chemical properties, soil organic matter, and soil biological processes [17,19]. In contrast, few research activities have treated the tillage effects on grain mineral elements, total phenolic content and total flavonoids content. This is despite of the tillage effects remarked on protein and gluten content [23], hormone activity [24] and sucrose content [19].

The main objective of this study was to determine the effect of tillage practices on some minerals elements uptake, total phenolic content and total flavonoid content of barley during tillering and grain filling stages in North West Tunisia under rainfed conditions.

2. MATERIALS AND METHODS

2.1 Trial Description

This trial was installed at the ESAK farm (Boulifa, Kef) situated in northwestern Tunisia (36°07'15.50"N; 8°43'24"E; altitude = 524 m). Boulifa is characterized by annual precipitation of about 427 mm. It had a Semi arid climate. The soil was alkaline (pH = 8.3) and sandy clay and relatively poor in organic matter (1.8%).

The trial have been installed since 2010-2011 growing season and the sampling was achieved in tillering and grain filling stages for the cultivation year 2016-2017. The biannual crop rotation was barley (Hordeum vulgare L.) cultivar ‘Rihane’ and durum wheat (Triticum durum L.) cultivar ‘Razzek’. Two tillage systems were tested: conventional tillage (CT) versus no-tillage (NT). For CT, reversible moldboard ploughing to 30-40 cm depth was applied followed by secondary tillage with offset 15-20 cm and a direct driller was used for NT plots. For NT, glyphosate (3 l.ha⁻¹) was applied to control weeds. The seeding rates of durum wheat and barley were respectively 140 kg.ha⁻¹ and 130 kg.ha⁻¹. At sowing, cultivation received 100 kg.ha⁻¹ of Di-Ammonium Phosphate. Then, ammonium nitrate (75 kg.ha⁻¹) was used at early tillering and (75 kg.ha⁻¹) at stem elongation stages.

2.2 Sampling and Measurements

For the cultivation years 2016-2017, a sampling was performed during tillering and grain filling stages. Aerial parts were dried, milled, sieved and stored for mineral contents, TPC and TFC.

2.3 Mineral Elements Determination

For mineral elements analysis, 1 g of dried sample were ashed in a muffle oven at 600 °C for 6h, and mineralized with HCl. Mineral elements concentrations were determined in quadruplicate. Potassium, calcium and sodium concentrations were estimated using the flame photometry. Spectrophotometry was used to determine phosphorus concentrations [25].

2.4 Extraction

Ground plant material (0.5 g) were put in 25 ml of methanol (80%) then shaken during 2 h and the solid phase was removed using a Whatman filter paper. For each treatment, four extracts were prepared and stored until analysis.

2.5 Determination of Total Phenolic Content (TPC)

A method based on Folin–Ciocalteu reagent, recommended by Singleton and Rossi [26] was used for TPC quantification. At 720 nm, the spectrophotometer was used to determinate absorbance of different extract and a blank after 1 h. Gallic acid (GA) was used for the standard curve (0–1000 ppm) and TPC was expressed as milligram of gallic acid equivalent (GAE) per gram of dry weight.

2.6 Estimation of Total Flavonoid Content (TFC)

The colorimetric method recommended by Zhishen et al. [27] and modified by Chaieb et al. [28] was used to measure TFC of durum wheat samples at 510 nm against a blank. Rutin was used for the calibration curve and TFC was expressed as milligram of rutin equivalents (RE) per gram of dry weight.

2.7 Statistical Analysis

The results were statistically analysed by Social Sciences software (SPSS 20.0, SPSS Inc., Chicago, IL, USA) to identify treatment effects and interactions (Two-way MANOVA and Pearson correlation). Duncan post hoc test was used to check differences between variables at the level of significance P = .05.
3. RESULTS AND DISCUSSION

3.1 Mineral Elements Contents

Minerals in plant serve to sustain many phytochemical processes. Macronutrients, such as phosphorus, calcium, and potassium are components of nucleic acids, proteins, hormones, phospholipids, coenzymes, adenosine triphosphate (ATP) and chlorophylls etc.

The plant contents of phosphorus, potassium, calcium, and sodium are shown in Fig. 1. Beyond the limitation of glyphosate use in no tillage and its effect on mineral content, analysis of variance revealed that tillage (T) had no significant effect on plant mineral composition in tillering stage neither in grain filling stage. However, higher contents of phosphorus, calcium, and potassium were noted under NT. Plant stage (S) presented significant effects on P, Ca, and Na contents. The interaction T x S had no significant effect on mineral elements contents.

These results are in accordance with those of Chaieb et al. [29] who studied the effect of tillage practices on nutrient uptake and demonstrated that P, K, Ca, and Na contents in durum wheat did not depend on tillage practices. Likewise, a study conducted by Ishaq et al. [30] revealed that P and K in wheat did not vary in relation to tillage practices. Besides, similar effects were noted for corn P and K concentrations [31].

In contrast, Chaieb et al [32] noticed a significant effect of tillage practices on durum wheat mineral contents at maturity stage under semi-arid conditions. In addition, corn mineral contents were affected by tillage and lower concentrations were noted under NT compared to CT. Guan et al. [34] studied maize mineral contents and found lower values for NT compared to CT.

As a result of fertilizer management, nutrients low movement and crop residue, NT causes nutrients accumulation in the top few centimetres of soil and wherefore, a reduction of the plant nutrients availability [35].

![Graphs showing mineral contents](image-url)
3.2 Total Phenolic Content (TPC)

Phenolic compounds have a main role in plant metabolism and plant defense system as they react against nematodes, fungi and insects attacks. As shown in Fig. 2, analysis of variance revealed that tillage did not affect significantly TPC. However, significant variability was noted for TPC according to growth stage and grain filling stage showed the highest TPC.

These results are in accordance with those of Chaieb et al. [32] and Chaieb et al. [36] who studied durum wheat TPC respectively at maturity stage and tillering stage as affected by tillage systems and found that no significant effects were noted. In similar, Stracke et al. [37] recorded that management operations did not influence wheat phytochemicals concentrations. In contrast, some studies reported that management practices influenced wheat TPC [38] and maize TPC [39].

3.3 Total Flavonoids Content (TFC)

In cereal, flavonoids are one of the major groups of phenolic compounds [40]. Thus, analysis of variance of TFC found that the effect of tillage is no significant. While, growth stage presented a significant effect on this parameter, and grain filling stage showed the highest TFC as shown Fig. 3.

These results are in accordance with those of Chaieb et al. [32] and Chaieb et al. [36] who studied durum wheat TFC respectively at maturity stage and tillering stage as affected by tillage systems and found that no significant effects were noted. In similar, Stracke et al. [37] recorded that management operations did not influence wheat phytochemicals concentrations. In contrast, Asami et al. [38] reported that management practices influenced wheat TFC.

3.4 Correlation among Mineral Contents, TPC and TFC

As shown in Table 1, phosphorus (P%) showed high positive significant correlation with Calcium (Ca%) and sodium (Na%) respectively \( r=0.902 \) and \( r=0.935 \) and high negative significant correlation with TPC and TFC respectively \( r=-0.710 \) and \( r=-0.638 \). Calcium (Ca%) showed significant correlation with sodium (Na%), TPC and TFC respectively \( r=0.949 \), \( r=-0.710 \) and \( r=-0.618 \). Although, significant correlations were noted among sodium (Na%) with TPC and TFC. Furthermore, a significant positive correlation was noted among TPC and TFC \( r=0.593 \). Partial correlations presented in Table 1 (a) and Table 1 (b) revealed that tillage system had the main part of these correlations.

![Fig. 2. Effect of conventional tillage (CT) and No tillage (NT) on TPC of barley in Boulifa during 2016-2017 cultivation year](image-url)
Fig. 3. Effect of conventional tillage (CT) and No tillage (NT) on TFC of barley in Boulifa during 2016-2017 cultivation year

Table 1. Correlation coefficients among mineral concentrations, total phenolic content and total flavonoids content of barley conventional tillage versus no tillage in Boulifa for 2016-2017 cultivation year

|          | P%  | K%  | Ca%  | Na%  | TPC  | TFC  |
|----------|-----|-----|------|------|------|------|
| P%       | 1   |     |      |      |      |      |
| K%       | .136| 1   |      |      |      |      |
| Ca%      | .902**| .199| 1    |      |      |      |
| Na%      | .935**| .220| .949**| 1    |      |      |
| TPC      | -.710**| -.150| -.710**| -.729**| 1    |      |
| TFC      | -.638**| -.124| -.618' | -.555' | .593' | 1    |

* Significant correlation p=.05.
** High Significant correlation p=.01.

a P%, phosphorus; K%, potassium; Ca%, calcium; Na%, Sodium; TPC, Total Phenolic Content; TFC, Total Flavonoids Content

Table 2. Partial correlation based on tillage practices (part a) and growth stages (part b) among mineral concentrations, total phenolic content and total flavonoids content of barley conventional tillage versus no tillage in Boulifa for 2016-2017 cultivation year.

Table 2. (Part a)

|          | P%  | K%  | Ca%  | Na%  | TPC  | TFC  |
|----------|-----|-----|------|------|------|------|
| P%       | 1   |     |      |      |      |      |
| K%       | .153| 1   |      |      |      |      |
| Ca%      | .902 | .219| 1    |      |      |      |
| Na%      | .935| .240| .949 | 1    |      |      |
| TPC      | -.750| -.096| -.753| -.771| 1    |      |
| TFC      | -.673| -.072| -.654| -.588| .566 | 1    |

* Significant correlation p=.05.
** High Significant correlation p=.01.

a P%, phosphorus; K%, potassium; Ca%, calcium; Na%, Sodium; TPC, Total Phenolic Content; TFC, Total Flavonoids Content
Table 2. (Part b)

|       | P%   | K%   | Ca%  | Na%  | TPC  | TFC  |
|-------|------|------|------|------|------|------|
| P%    | 1    |      |      |      |      |      |
| K%    | -1.138 | 1   |      |      |      |      |
| Ca%   | -1.198 | 0.037 | 1   |      |      |      |
| Na%   | 0.378   | 0.115 | 0.240 | 1   |      |      |
| TPC   | 0.033   | -0.001 | 0.272 | 0.011 | 1   |      |
| TFC   | -0.026  | 0.010   | 0.258 | 0.409 | 0.164 | 1   |

*a Significant correlation p=.05. **High Significant correlation p=.01.

These results are similar to those of Chaieb et al. [29] who found that P% had positive correlation with K% and that based on tillage practices P% presented positive significant correlations with Ca% and Na% for durum wheat during tillering stage. However, durum wheat grain had not presented any significant correlations for these parameters [32].

4. CONCLUSION

This work is a contribution to discern the effect of tillage on mineral elements uptake, total phenolic content and total flavonoid content in relation to growth stage. The results revealed that under rainfed conditions in North West Tunisia, plant mineral uptake were more affected with plant stage than tillage practice. Moreover, tillage had no significant effect on total phenolic content and total flavonoids content which presented significant variability according to plant stage. These results should encourage farmers in North West Tunisia to adopt No tillage for barley cultivation. These practices permit to limit soil erosion in this region and enhance the sustainability of cereal production.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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