



The effect of iron and zinc fertiliser dosage on yellow rust in durum wheat (*Triticum durum* L.)

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Accepted: 27 May 2025 / Published online: 9 June 2025
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Abstract This study aimed to assess the impact of dosage of iron (Fe) and zinc (Zn) on the incidence and severity of yellow rust (*Puccinia striiformis* f. sp. *tritici*) in durum wheat (*Triticum durum*), utilizing vegetation indices derived from multispectral data for precise analysis. For this purpose, freshly collected yellow rust spores were homogenized in volatile minerals and applied to all durum test materials (Kızıltan 91, Eminbey, Mirzabey 2000, Çeşit-1252) under artificial epidemic conditions in two periods, heading (Zadoks 45) and flowering (Zadoks 50), one week apart. In addition, each of the blocks, which were arranged in three replicates and four durum wheat cultivars, received different doses of iron (Fe), zinc (Zn) and Fe+Zn. Mineral fertilizers were applied in six phenological periods (Zadoks13-Zadoks21, Zadoks 22-Zadoks37 and Zadoks41-Zadoks49) starting from the tillering period (Zadoks 13-21). The

effects of mineral fertilizer applications on the disease were evaluated by converting the reflectance values obtained from multispectral data into vegetation index values for each phenological period. Iron (Fe) applications used for durum cultivars decreased disease severity in the early period (Zadoks 13-21) but increased it in the late period (Zadoks 41-49). Zinc (Zn) applications reduced disease severity in early and late phenological growth periods. Iron (Fe)+ Zinc (Zn) applications reduced disease severity in all phenological periods. The vegetation indices for monitoring disease severity were LCCI, SIPI, PSRI, NDVI, NBNDVI, MSR, GNDVI, NLI, and YRI (Table 1). These indices were used for disease detection for all phenological periods. Iron, zinc and iron+zinc applications resulted in a decreasing disease severity reflected in the GNDVI and LCCI indices in the early-mid period (Zadoks17), and in the NBNDVI and LCCI indices for all other durum varieties.

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Keywords Wheat · Hyperspectral data · Phenological period · Yellow rust · Disease severity · Mineral fertilizer

Introduction

Control of diseases and pesticides in wheat is important for good yields. In this respect, it has become imperative to apply the least harmful methods to human health and the environment to

increase yield and quality and minimize stress factors (Atkinson & McKinlay, 1997; Batish et al., 2007; Camprubí et al., 2007). It is important to provide the nutrients needed by plants and microorganisms in sufficient quantities and at the appropriate timing to control diseases (Agrios, 2005). Rust diseases caused by *Puccinia* spp. significantly reduce yield and quality of wheat (Chen, 2020; Jørgensen et al., 2024; Tekin et al., 2022). Rust diseases wind dispersed. Rusts threaten wheat production globally (Riella et al., 2024; Tekin et al., 2021).

To monitor the development of disease at different crop growth stages, the relationships between spectral reflectance values and biophysical characteristics of the plants have been examined and regression models were developed to represent these relationships. The optimal yield-related spectral indices and their combinations were found. Hyperspectral indices are used to distinguish between diseased and stress-affected plants (Moshou et al., 2004). There are many vegetation indices developed for the canopy-level diagnosis of rust diseases and septoria leaf spot disease in winter wheat (Liu et al., 2020; Heidarian Dehkordi et al., 2020). In addition, broadband vegetation indices are already in use to distinguish between diseased and healthy plants (Reis Pereira et al., 2024).

In a study by Huang et al. (2007) in the seedling stage (greenhouse) and the adult plant stage (field), the relationship between the Photochemical Reflectance Index (PRI) and disease was investigated (Ganeva et al., 2024) and a significant correlation was found. In a study by Devadas et al., 2009, ten vegetation indices (VIs) were used to differentiate yellow rust, leaf rust, and stem rust. The, Anthocyanin Reflectance Index (ARI) and the Transformed Chlorophyll Absorption Reflectance Index (TCARI) were found to be effective in disease discrimination.

Micronutrients effect phenol and lignin content and membrane stability in plants. In a study conducted by Graham and Webb (1991), it was revealed that the use of Manganese (Mn) and Zinc (Zn) instead of fungicides in disease prevention is an alternative that gives effective disease control without causing environmental pollution. The role of zinc (Zn) in plant growth is mediated by auxin (Ohki, 1978), which enhances disease resistance and increases meristematic activity, leading to greater cell division and expansion. Moreover, the use of iron (Fe), Mn, and Zn increases the

amount of these elements in the grain and flag leaf (Potarzycki & Grzebisz, 2009).

While the physiological roles of plant nutrients are generally well established, the interactions between micronutrients and phytopathogens are still not fully understood and require further investigation. Recent studies have shown that timely and adequate fertilizer application is important for the control of some phytopathogens (Graham & Webb, 1991 micronutrients; Marschner, 1995; Huber & Graham, 1999).

Some recent studies have revealed that nitrogen (N), phosphorus (P), potassium (K), manganese (Mn), zinc (Zn), boron (B), silicium (Si), chlorine (Cl) applications are effective in reducing disease severity. The general view in reducing plant diseases has been that nutrient applications can be reduced to an acceptable level (Nadeem et al., 2018). The ability of to reduce disease is related to the physiology and biochemistry of the plant. Each has important responsibilities in reducing disease severity and controlling pathogens. Although iron (Fe) is an important element in human health, its effect on plants has not been fully analyzed. While higher plants require high levels of iron for growth, it has been found effective in reducing the severity of foliar diseases in plants such as wheat and banana (Graham, 1983; Graham & Webb, 1991). Zinc (Zn) had a disease-reducing effect on some foliar diseases but had no positive effect on others (Graham & Webb, 1991). Zinc may reduce disease severity through its direct toxic effect on the pathogen, while having no adverse impact on the plant's metabolism (Graham & Webb, 1991).

This study aimed to investigate the changes in the electromagnetic spectrum (331–1141 nm) of some durum wheat cultivars exposed to artificial rust (caused by *Puccinia striiformis* f. sp. *tritici*) under mineral fertilizer (Fe, Zn, Fe + Zn) applications.

Materials and methods

Climate and soil characteristics of the experimental site

The research was done during the 2018–2019 production season at the experimental field of the Field Crops Central Research Institute, located in the Yenimahalle district of Ankara province. The experimental area received an average rainfall of 367 mm from

September to August, with an average temperature of 12.08 °C (Anonymous, 2019). The soil in the experimental field was characterized as clayey-loamy in texture.

Plant materials

The durum wheat cultivars Eminbey (resistant), Kızıltan 91 (moderately susceptible), Çeşit-1252 (moderately susceptible), and Mirzabey 2000 (moderately susceptible) (Aydoğdu & Akan, 2023; Ipek et al., 2023), which are known to respond to fertilizer applications and were selected as experimental material.

To measure the distribution of the disease in the production area and to control its development, “Little Club” was planted between the replicates as a susceptible control genotype.

Fertilizer doses and experimental design

Durum wheat experimental materials was sown by hand on November 14 th, 2018 with 4 rows and 2.5–3 g of seeds per row. Diammonium phosphate fertilizer (DAP) was applied as 6.3 g/0.45 m² for 14 kg per decare. Iron (Fe), Zinc (Zn), and Iron + Zinc (Fe + Zn) mineral fertilizer doses were applied to each of the replicates with a width of 0.60 m and a length of 10 m in 6 phenological periods (Zadoks13–Zadoks21, Zadoks22–Zadoks37 and Zadoks41–Zadoks49) starting from the tillering period (Zadoks13–21).

For iron (Fe) dose application, 75–100 g Fe-6 Forte® was dissolved in 100 l of water and applied to 1 decare. Accordingly, three different fertilizer application doses for Fe; 5 g/5 lt/6 m² Fe in the first application, 10 g/5 lt/6 m² Fe in the second application, and 20 g/5 lt/6 m² Fe in the third application were prepared and applied.

For zinc (Zn) dose application, Powder-Forte® form was prepared for 1 decare by dissolving 150 g in 100 l of water (0.60 m*10 m = 6 m²). It was prepared and applied as 7.5 g/5 L/6 m² Zn in the first application, 15 g/5 L/6 m² Zn in the second application, and 30 g/5 L/6 m² Zn in the third application.

For the iron (Fe) + Zinc (Zn) dose application, Fe 5 g/5 lt + Zn 7.5 g/5 lt was used in the first application, Fe 10 g/5 lt + Zn 15 g/5 lt in the second application, and Fe 20 g/5 lt + Zn 30 g/5 lt in the third

application starting from the tillering period. The applications were repeated in a total of 6 periods.

Inoculation process and disease evaluation

Fresh disease spores used in the study were obtained from the national Bitki Protection Standards (Li et al., 1989). Disease spores were inoculated into wheat materials when there was no wind and the sun was close to setting. The disease spores were homogenized in essential mineral oil (Soltrol 170®) at 2 mg/100 ml. and applied to all replicates (I + II + III) with ULV + device on 2 different dates, the first one was 172 days after sowing at emergence (06 May 2019) and the second one was 179 days after sowing at pre-flowering (13 May 2019). To prevent spore transfer to neighboring plots, inoculation plots were closed to spore passage with plastic polycarbonate barriers.

Yellow rust samples were taken on Zadoks stage 41–43–46 and 49. These periods were the beginning of flowering (Zadoks 60), grain filling (Zadoks 63), milk maturity (Zadoks 71), and yellowing (Zadoks 75).

The Modified Cobb scale was used to determine yellow rust reactions, primarily the disease indices (DI%) (Peterson et al., 1948) the type of plant reaction to disease and the corresponding Coefficient Infection (R:0.2; MR:0.4; MR-MS:0.6; MS:0.8; S:1) (Roelfs et al., 1992). The disease severity (DS%) value was obtained by multiplying the disease index (DI%) value calculated for each of the test materials by the corresponding infection Coefficient Infection (CI) value (Peterson et al., 1948).

As a result, 5 yellow rust classes were created according to the severity value by Akan (2019). These yellow rust reaction groups were classified as IC = 0; Immune, IC ≤ 0.1–5; Resistant, CI ≤ 5.1–20; Moderate Resistant, CI ≤ 20.1–40; Moderate susceptible, IC ≤ 40.1–100; Susceptible, IC = 0–20; Resistant group, IC ≤ 20.1–100; susceptible group.

Processing of images and classification

To determine the average disease severity, an average disease score was calculated for each cultivar from each replicate, and based on this score, an average disease severity (DS%) was calculated for each cultivar. The ‘Image Classification’ module in ArcGIS

10.5.1® was used to assign a disease score to each cultivar through the Unsupervised Classification application. Disease severity (DS%) was determined by calculating the ratio of the leaf area affected by yellow rust to the total leaf area, and then multiplying this ratio by an efficiency coefficient. Based on disease severity, plants were categorized into nine distinct groups for disease evaluation: 0%, 1%, 10%, 20%, 30%, 45%, 60%, 80%, and 100%. A score of 0% indicated the absence of disease on the leaf, while a score of 100% represented the most severe disease condition, where the leaf was completely covered by the infection. The disease index was calculated using the formula provided by Huang et al. (2007) [1]. The final disease severity (DS%) was obtained by multiplying the disease index by the Coefficient of Infection, which incorporates the type of reaction [2].

$$DI(\%) = \frac{\sum xf}{n \sum f} \times 100 \quad (1)$$

DI (%) = Disease Indice.

f = Number of leaves at each degree of disease severity.

n = Highest disease severity value.

x = intensity at each disease level

$$DS(\%) = DI(\%) \times CI \quad (2)$$

DS (%) = Disease Severity.

DI (%) = Disease Indice.

CI = Coefficient of Infection.

Scoring of diseases using an image classification technique

In the classification technique used for numerical evaluation of the disease, 5 leaf samples were randomly selected from each cultivar and photographed with a digital and thermal camera. The colour co-occurrence method (RGB “Red - Green-Blue”) of diseased colour co-occurrence leaf images acquired with a thermal camera was used to classify diseased leaf images using ArcGIS. 10.5 Programme® “Image Classification Module”. First, eight class files were created to identify the diseased regions of the image. Green areas were masked and diseased areas (pustules) were identified from this file. According to the number of pixels, the area covered on the leaf surface was calculated as a percentage.

Spectral mixture analysis (SMA) was used to determine the disease score by determining the different colour ratios in diseased areas compared to healthy plant leaves (Franke & Menz, 2007). For this purpose, using the ArcGIS 10.1® Image Classification Unsupervised technique, the colour symptom ratio of the remaining diseased areas was determined by masking the green areas with the unsupervised classification technique at the pixel level and the “diseased area score” was determined. The severity of the disease (DS%) was determined by multiplying the determined disease score by the corresponding infection (CI) value.

Spectral reflectance measurements of durum wheat

Spectroradiometric measurements of canopy reflectance were made with a hand-held portable spectroradiometer between 11:00 and 15:00 when there was no cloud cover and the sunlight was perpendicular to the ground. Spectral reflectance measurements were conducted at weekly intervals using a spectral sensor with a wavelength of 330–1150 nm, captured at 3 nm intervals, at different stages of wheat development, from tillering to milking. Measurements were taken at a 25° angle to the ground and from a height of 25 cm above the plant crown. For plant spectral reflectance measurements, “3 values” (3 replicates * 3 values) were taken for each replicate in the three rows of each cultivar for which a disease response was observed. The result was a total of 9 reads and a total of 36 reads for 4 durum cultivars (4 cultivars * 9 readings). The same protocol was applied to durum cultivars and a total of 36 measurements were made.

Calculation of vegetation indices and statistical analyses

For the diagnosis of wheat yellow rust, Table 1 shows the vegetation indices calculated based on the reflectance values of the electromagnetic spectrum band 331–1141 nm. The correlation between disease severity values (DS%) and vegetation indices values were evaluated. The correlation values were used to develop regression models for disease response prediction.

To test the performance of the disease prediction regression models, the correlation between observed disease responses (observed actual disease estimates) and vegetation indices were calculated. In addition

Table 1 Vegetation indices that differentiate between diseased and healthy plants

Vegetation Indices	Explanation	Formula	Region of Influence
NDVI	Normalized Difference Vegetation Index	$NDVI = (R_{830} - R_{675}) / (R_{830} + R_{675})$	Leaf Area Index, Photosynthetically Active Radiation (PAR), or Biomass (PAB)
NBNDVI	Narrow-Band Normalized Difference Vegetation Index	$NBNDVI = (R_{850} - R_{680}) / (R_{850} + R_{680})$	Used to Follow Vegetation
NRI	Nitrogen Reflectance Index	$NRI = (R_{570} - R_{670}) / (R_{570} + R_{670})$	Nitrogen Status
PRI	Photochemical Reflectance Index	$PRI = (R_{570} - R_{531}) / (R_{570} + R_{531})$	Photosynthetic Radiation (PR)
TCARI	The Transformed Chlorophyll Absorption and Reflectance Index	$TCARI = 3 \times [(R_{700} - R_{675}) - 0.2 \times (R_{700} - R_{550}) \times (R_{700} / R_{670})]$	Chlorophyll a + b Concentration
SIPI	Structural Independent Pigment Index	$SIPI = (R_{800} - R_{445}) / (R_{800} - R_{680})$	Pigment Concentrations
PhRI	Physiological Reflectance Index	$PhRI = (R_{550} - R_{531}) / (R_{550} + R_{531})$	Calculating Light Usage Efficiency
NPCI	Normalized Pigment Chlorophyll Ratio Index	$NPCI = (R_{680} - R_{430}) / (R_{680} + R_{430})$	Calculation of Pigment Coverage
ARI	Anthocyanin Reflectance Index	$ARI = (R_{550})^{-1} - (R_{700})^{-1}$	Anthocyanin Content
CARI	Chlorophyll Absorption Ratio Index	$CARI = ([(a670 + R_{670} + b)] / (a2 + 1) / 2) \times (R_{700} / R_{670})$ $a = (R_{700} - R_{550}) / 150$, $b = R_{550} - (a \times 550)$	Calculating Chlorophyll Absorption
GI	Green Index	$GI = R_{554} / R_{677}$	Calculation of Pigment Coverage
TVI	Triangular Vegetation Index	$TVI = 0.5[120(R_{750} - R_{550}) - 200(R_{670} - R_{550})]$	Determination of Plant Status
MCARI	Modified Chlorophyll Absorption in Reflectance Index	$MCARI = (R_{701} - R_{671}) - 0.2(R_{701} - R_{549}) / (R_{701} / R_{671})$	Calculating Chlorophyll Absorption
PSRI	Plant Senescence Reflectance Index	$PSRI = (R_{680} - R_{500}) / R_{750}$	Pigment Coverage, Leaf Maturation, and Yellowing
RVSI	Red-Edge Vegetation Stress Index	$RVSI = [(R_{712} + R_{752}) / 2] - R_{732}$	Internal Structure Parameters
WI	Water Index	$WI = R_{900} / R_{970}$	Calculation of Changes in Water Amount
LCCI	Leaf and Canopy Chlorophyll Index	$LCCI = (R_{750} - R_{705}) / (R_{750} + R_{705})$	Calculating Chlorophyll Absorption
NVI	New Vegetation Index	$NVI = (R_{777} - R_{747}) / (R_{673})$	Calculating Chlorophyll Absorption
GNDVI	Green Normalized Difference Vegetation Index	$GNDVI = (R_{750} - R_{550}) / (R_{750} + R_{550})$	Leaf Area Index, Photosynthetically Active Radiation (PAR), or Biomass (PAB)
SRPI	Simple Ratio Pigment Index	$SRPI = R_{430} / R_{680}$	Calculation of Pigment Coverage
RVI	Ratio Vegetation Index	$RVI = R_{NIR} / R_{Red}$	Biophysical Changes and Disease Monitoring
RDVI	Renormalized Difference Vegetation Index	$RDVI = (R_{800} - R_{670}) / (R_{800} + R_{670})^{0.5}$	Relationships Between Biophysical Parameters
DVI	Difference Vegetation Index	$DVI = R_{890} - R_{670}$	Investigation of Changes in Vegetation
NLI	Non-Linear Vegetation Index	$NLI = (R_{NIR}^2 / R_R) / (R_{NIR}^2 + R_R)$	Leaf Area Index Calculation
SR	Simple Ratio	$SR = R_{NIR} / R_R$	Monitoring Vegetation
MSR	Modified Simple Ratio Index	$MSR = (R_{800} / R_{670} - 1) / \sqrt{R_{800} / R_{670} + 1}$	Leaf Area Index Calculation
YRI	Yellow Rust Index	$YRI = (R_{730} - R_{419}) / (R_{730} + R_{419}) + 0.5R_{736}$	Monitoring Yellow Rust

to this calculation, the “Root Mean Squared Error (RMSE)” and “Standard Error (SE%)” values were calculated separately for each index. As a result, it

was found that using higher R^2 values and lower RMSE and SE% indices gave effective results for determining plant disease severity (DS%).

Basic statistics, variance, correlation, and regression analysis for all phenological stages were performed using the SPSS-22® version of the statistical package (IBM SPSS Statistics, 2014).

Root Mean Square Error (RMSE) [3]) and Percentage Relative Error (RE%) [4]) are calculated using the following equation.

$$\text{Root Mean Square Error (RMSE)} = \sqrt{\frac{1}{n} \sum_{i=1}^N (\bar{y}_i - \hat{y}_i)^2} \quad (3)$$

In the formula:

$\hat{y}_i$ = Calculated value from Correlation Equation.

$\bar{y}_i$ = Average Disease Severity Value (%DS).

n = Number of Samples Used in the Experiment.

$$\text{Relative error percentage (RE\%)} = \frac{\text{Root Mean Square Error (RMSE)}}{\bar{y}_i} * 100 \quad (4)$$

Results

The effects of different mineral fertilizers on yellow rust responses of durum wheat cultivars

In the Kızıltan 91 cultivar, mineral fertilizer applications (Fe, Zn) were effective in increasing the disease severity in the early-middle and middle-late period, and Fe + Zn fertilizer dose applications were effective in decreasing the disease severity in the middle-late period (Table 2). In Fe fertilizer applications, the SIPI, PSRI, NDVI, RDVI, NBNDVI and LCCI indices had a high correlation with

the disease severity indice in the mid-late period. In the Zn fertilizer applications, the MCARI, NPCI, SRPI, PRI, and TCARI indices had a high correlation with the disease indice in the early-mid period. In the Fe + Zn fertilizer applications, high correlation values were found in the NBNDVI, NVI, NDVI, GNDVI, and MSR indices in the late period in the direction of decreasing disease severity.

High correlation values were determined in Fe and Zn fertilizer doses of mineral fertilizer applications in all phenological periods in the Çeşit-1252 cultivar (Table 3), and the effect of mineral fertilizer

Table 2 Monitoring the effects of different phenological periods of the Kızıltan 91 cultivar and different mineral fertilizer dose applications on yellow rust reactions by vegetation indices

Fertilizer Application	Effective Period	Vegetation Indices (VI)	Band Range (nm)	Correlation (R ²)	RMSE*	%SE**	Regression Eq. (DI)
Fe5	10.5.4 (+)	SIPI	800-445	0.768	3.839	10.664	y = 0.5906x + 1.529
Fe10		PSRI	750-500	0.756	3.928	10.912	y = 0.5713x + 1.6004
Fe20		NDVI	830-675	−0.696	4.306	11.962	y = 0.4848x + 1.9237
		NBNDVI	850-680	−0.696	4.309	11.969	y = 0.4842x + 1.9254
		LCCI	750-705	−0.694	4.317	11.992	y = 0.4822x + 1.9334
Zn7.5	10.5.3 (+)	MCARI	701-549	0.849	3.172	8.812	y = 0.7204x + 1.0435
Zn15		NPCI	680-430	0.823	3.411	9.476	y = 0.6767x + 1.207
Zn30		SRPI	845-770	−0.809	3.530	9.805	y = 0.6539x + 1.2924
		PRI	570-531	0.763	3.880	10.777	y = 0.5819x + 1.5608
		TCARI	700-550	0.740	4.037	11.213	y = 0.5474x + 1.6902
Fe5 + Zn7.5	10.5.4 (−)	NBNDVI	850-680	0.819	1.395	3.874	y = 0.6714x + 3.0669
Fe10 + Zn15 Fe20 + Zn30		NVI	777-673	0.771	1.549	4.302	y = 0.5948x + 3.7824
		NDVI	830-675	0.755	1.596	4.434	y = 0.5696x + 4.0165
		GNDVI	750-550	0.746	1.621	4.504	y = 0.5559x + 4.1457
		MSR	800-670	0.717	1.696	4.710	y = 0.5143x + 4.5335

applications on the reduction of disease severity was detected. In Fe fertilizer treatments, the SRPI, NPCI, WI, DVI, and TVI indices had a high correlation with the disease severity in the early-mid period. In Zn fertilizer treatments, the PhRI, TCARI, MCARI, PRI, and GNDVI indices in the 531–750 nm band range in the early period ($R^2 = 0.671\text{--}0.833$), LCCI, MSR, NDVI, SIPI and PSRI indices had a high correlation with disease severity in the middle-late period.

In the Eminbey cultivar, Fe and Zn fertilizer applications showed high correlation between the vegetation indices and disease severity (Table 4). Fe fertilizer reduced disease severity in the early-mid period, and Zn fertilizer application doses reduced disease in the early-midperiod. In Fe + Zn fertilizer applications, it was determined that the LCCI, GNDVI, SIPI, NBNDVI, and PSRI indices showed high correlation values with disease severity in the early-mid period. Among the mineral fertilizer applications, Fe and Zn fertilizer dose applications showed high correlation values with disease severity in all phenological periods.

In the Mirzabey cultivar (Table 5). Fe fertilizer dose applications decreased disease severity in the early-mid period, in the mid-late period in Zn fertilizer dose applications. In Fe + Zn fertilizer dose applications, showed high correlation values with disease severity ($R^2 = 0.938\text{--}0.961$).

Spectral band regions and indices effective in monitoring disease severity

When durum cultivars were evaluated together

The Eminbey cultivar in the early-middle period in the Red + NIR, and the Mirzabey 2000 cultivar in the middle-late period in the Green + Red region showed an increase in disease severity. The Kızıltan 91 cultivar in the early period, and the Çeşit-1252 cultivar in the middle-late period, in the Red + NIR region showed high correlations with the decrease in disease severity. For all durum cultivars except the Mirzabey 2000, the effective band region is located in the RED + RED EDGE + NIR range, and increases in disease severity values were determined. The effective band region for the Mirzabey 2000 cultivar was determined as Green + Red. LCCI, SIPI, PSRI, NDVI, NBNDVI, MSR, GNDVI, NLI, and YRI were the prominent indices in the reaction evaluation of durum cultivars (Table 6) (Aydoğdu et al., 2023).

The vegetation indices for the determination of disease severity according to phenological periods vary in fertilizer applications for durum cultivars. The SIPI, PSRI, and LCCI indices may be effective in determining disease severity in the middle-late

Table 3 Monitoring the effects of different phenological periods of the Çeşit-1252 cultivar and different mineral fertilizer dose applications on yellow rust reactions by vegetation indices

Fertilizer Application	Effective Period	Vegetation Indices (VI)	Band Range (nm)	Correlation (R^2)	RMSE*	%SE**	Regression Eq. (DI)
Fe5	10.5.3	SRPI	845-770	-0.863	0.815	2.263	$y = 0.7445x + 2.6116$
Fe10	(-)	NPCI	680-430	0.857	0.830	2.306	$y = 0.7346x + 2.7129$
Fe20		WI	900-970	0.687	1.171	3.254	$y = 0.4718x + 5.4002$
		DVI	890-670	0.684	1.176	3.267	$y = 0.4673x + 5.4439$
		TVI	750-550	0.604	1.285	3.570	$y = 0.3628x + 6.484$
Zn7.5	10.5.1	PhRI	550-531	0.833	3.813	10.593	$y = 0.6934x + 0.6947$
Zn15	(-)	TCARI	700-550	0.827	3.874	10.760	$y = 0.6836x + 0.7171$
Zn30		MCARI	701-549	0.793	4.192	11.643	$y = 0.6295x + 0.8396$
		PRI	570-531	0.730	4.710	13.083	$y = 0.5323x + 1.0599$
		GNDVI	750-550	-0.671	5.107	14.185	$y = 0.4501x + 1.246$
Fe5 + Zn7.5	10.5.4	LCCI	750-705	0.755	0.507	1.408	$y = 0.5703x + 5.5378$
Fe10 + Zn15	(-)	MSR	800-670	0.754	0.508	1.412	$y = 0.5679x + 5.5698$
Fe20 + Zn30		NDVI	830-675	0.754	0.508	1.412	$y = 0.5681x + 5.5666$
		SIPI	800-445	-0.754	0.508	1.411	$y = 0.5687x + 5.5598$
		PSRI	750-500	-0.754	0.508	1.412	$y = 0.568x + 5.5675+$

Table 4 Monitoring the effects of different phenological periods of the Eminbey cultivar and different mineral fertilizer dose applications on yellow rust reactions by vegetation indices

Fertilizer Application	Effective Period	Vegetation Indices (VI)	Band Range (nm)	Correlation (R^2)	RMSE*	%SE**	Regression Eq. (DI)
Fe5	10.5.3	LCCI	750-705	0.792	1.680	4.666	$y = 0.6265x + 4.4822$
Fe10	(-)	TVI	750-550	0.779	1.723	4.786	$y = 0.6067x + 4.713$
Fe20		RDVI	800-670	0.773	1.744	4.845	$y = 0.5974x + 4.8311$
		RVSI	752-712	0.759	1.791	4.975	$y = 0.5755x + 5.0944$
		GNDVI	750-550	0.737	1.857	5.157	$y = 0.5438x + 5.4748$
Zn7.5	10.5.3	NVI	777-673	0.786	2.754	7.649	$y = 0.6184x + 6.1047$
Zn15	(-)	GNDVI	750-550	0.767	2.859	7.942	$y = 0.5887x + 6.5808$
Zn30		LCCI	750-705	0.763	2.882	8.006	$y = 0.5821x + 6.6872$
		MSR	800-670	0.741	2.995	8.320	$y = 0.5486x + 7.222$
		RDVI	800-670	0.733	3.033	8.426	$y = 0.5371x + 7.4073$
Fe5 + Zn7.5	10.5.3	LCCI	750-705	0.902	1.927	5.353	$y = 0.3366x + 8.5505$
Fe10 + Zn15	(-)	GNDVI	750-550	0.857	1.219	3.386	$y = 0.7346x + 3.4217$
Fe20 + Zn30		SIPI	800-445	-0.845	1.264	3.510	$y = 0.7148x + 3.6755$
		NBNDVI	850-680	0.833	1.309	3.637	$y = 0.6937x + 3.9478$
		PSRI	750-500	-0.827	1.330	3.695	$y = 0.6838x + 4.0755$

Table 5 Monitoring the impact of various phenological stages of the Mirzabey 2000 cultivar and different mineral fertilizer dosages on yellow rust reactions using vegetation indices

Fertilizer Application	Effective Period	Vegetation Indices (VI)	Band Range (nm)	Correlation (R^2)	RMSE*	%SE**	Regression Eq. (DI)
Fe5	10.5.3	DVI	890-670	0.853	1.674	4.650	$y = 0.7271x + 8.3697$
Fe10	(-)	NVI	777-673	0.818	1.845	5.124	$y = 0.6686x + 10.162$
Fe20		GNDVI	750-550	0.808	1.888	5.245	$y = 0.6529x + 10.646$
		NBNDVI	850-680	0.808	1.887	5.242	$y = 0.6533x + 10.635$
		LCCI	750-705	0.796	1.939	5.387	$y = 0.6338x + 11.231$
Zn7.5	10.5.4	NLI	845-770	-0.797	2.149	5.970	$y = 0.6347x + 10.876$
Zn15	(-)	WI	970-900	0.789	2.183	6.064	$y = 0.6231x + 11.219$
Zn30		YRI	736-419	0.687	2.584	7.177	$y = 0.4721x + 15.719$
		DVI	890-670	0.672	2.634	7.317	$y = 0.4514x + 16.337$
		RVSI	752-712	-0.646	2.714	7.538	$y = 0.4176x + 17.343$
Fe5 + Zn7.5	10.5.1	LCCI	750-705	-0.961	1.839	5.109	$y = 0.9233x + 0.9624$
Fe10 + Zn15	(-)	SIPI	800-445	0.959	1.873	5.202	$y = 0.9204x + 0.9939$
Fe20 + Zn30		PSRI	750-500	0.950	2.071	5.752	$y = 0.9028x + 1.2182$
		GNDVI	750-550	-0.940	2.259	6.276	$y = 0.8842x + 1.4503$

period in Fe fertilizer applications, the MCARI and PRI indices may be useful in determining disease severity in the early-middle period in Zn fertilizer applications, and NBNDVI, NDVI, and GNDVI indices may be effective in determining disease severity in Fe + Zn fertilizer applications in the middle-late

period in the Kızıltan 91 cultivar. It was determined that the use of DVI indices in Fe fertilizer applications in the early-mid period, the MCARI, PRI, and GNDVI indices in Zn fertilizer applications in the early period, and the LCCI, NDVI, SIPI and PSRI indices in Fe + Zn fertilizer applications in the

Table 6 Effective spectral band regions and indices in the monitoring disease severity (DS%) according to phenological periods of different fertilizer dosage applications in durum wheat cultivars

Cultivars	Fertilizer	R ²	Band Range (nm)	Effective Band Region	Effective Period	DS%	Effective Indices	Correlation Range (R ²)
Kızıltan 91	Fe	0.507	719–912	Red Edge + NIR	Early *	–	SIPI, PSRI, NDVI, NBNDVI, LCCI	0.768-0.694
Çeşit-1252	Fe + Zn	0.621	685–855	Red + NIR	Mid-Late***	–	LCCI, MSR, NDVI, SIPI, PSRI	0.755-0.754
Eminbey	Fe + Zn	0.573	692–871	Red + Red Edge + NIR	Early-Mid**	+	LCCI, GNDVI, SIPI, NBNDVI, PSRI	0.902-0.827
Mirzabey 2000	Zn	0.632	547–692	Green + Red	Mid-Late***	–/+	NLI, WI, YRI, DVI, RVSI	0.797-0.646

* (–) Early Term (25 May 2019)

** (+) Early-Mid Term (06 June 2019)

*** (–/+) Mid-Late Term (15 June 2019)

Table 7 Effective band areas and indices for disease determination in fertilizer application doses according to phenological periods

Application	Phenological Period*	Band Range	Band Region	Effective Indices
Fe	Early	692–912	Red + Red Edge + NIR	–
	Early-Middle	689–1016	Red + RedEdge + NIR	DVI, LCCI, GNDVI (–)**
	Middle-Late	419–726	Blue + Green + Red + Red Edge + NIR	SIPI, PSRI, LCCI (+)*
	Late	510–1035	–	–
Zn	Early	712–890	Red + Red Edge + NIR	MCARI, PRI, GNDVI (–)**
	Early-Middle	689–987	Red Edge + NIR	MCARI, PRI (+), GNDVI (–)**
	Middle-Late	–	–	–
	Late	675–921	–	–
Fe + Zn	Early	695–887	Red + Red Edge + NIR	SIPI, PSRI, GNDVI (–)**
	Early-Middle	692–871	Red Edge + NIR	LCCI, GNDVI, SIPI, NBNDVI (–)**
	Middle-Late	685–855	Red + Red Edge + NIR	NBNDVI, NDVI, GNDVI, LCCI, NDVI, SIPI, PSRI (–)**
	Late	685–855	–	–

*(+): Increase in disease severity

**(–): Decrease in disease severity

Phenological Periods*

10.5.1: Early Period (Beginning of Flowering)

10.5.3: Early-mid Period (Grain Filling Period)

10.5.4: Mid-late Period (Milking Period)

11.1: Late Period (Yellowing)

mid-late period may be effective in determining disease severity in cultivar-1252. In the Eminbey cultivar, the LCCI and GNDVI indices in Fe fertilizer applications in the early-mid period decreased the disease severity, the GNDVI index in Zn fertilizer applications, and the LCCI, GNDVI, SIPI and NBNDVI indices in Fe + Zn fertilizer applications in the early-mid period are effective in determining disease severity. In the Mirzabey 2000 cultivar, the DVI and GNDVI indices in Fe fertilizer applications in the early-mid period decreased the disease severity, the NLI indices in Zn fertilizer applications in the middle-late period decreased the disease severity, and LCCI, SIPI, PSRI and GNDVI indices in Fe + Zn fertilizer applications in the early period may be effective in decreasing the disease severity Table 7 (Aydoğdu et al., 2023).

Discussion

Many spectral indices have recently been developed to identify and distinguish crop host diseases. In a study by Huang et al. (2007), vegetation indices, the Yellow Rust Index (YRI), are proposed to distinguish yellow rust from healthy wheat, powdery mildew, and aphids using canopy and field surveys. Zhang et al. (2012) determined that the Physiological Reflectance Index (PhRI) effectively detected yellow rust across all stages of plant growth. Devadas et al. (2009) reported that at the leaf scale, no reflectance index can fully differentiate rust diseases (yellow rust, leaf rust, and stem rust). However, the Anthocyanin Reflectance Index (ARI) can distinguish diseased from healthy plants.

Many micronutrients have been reported to have distinct and important roles in inducing resistance or tolerance responses to pathogens. Iron (Fe) is an essential element for plant health. However, there are few studies on the effects of Fe applications on plant disease resistance or tolerance. Iron can reduce disease severity or control disease to varying degrees in many foliar diseases such as wheat and banana rusts (Graham, 1983; Graham & Webb, 1991). Zinc can cause different responses to disease in sensitive plant responses. Although some applications may reduce disease severity and

incidence, some applications may not (Graham & Webb, 1991). In many cases, the application of Zn can reduce disease severity due to its direct toxic effect on the pathogen but not on plant metabolism (Graham & Webb, 1991).

As a result of our study, it was possible to identify infected and non-infected leaves with 70% accuracy using vegetation indices. Moshou et al. (2004) reported that the most sensitive bands in the spectral range that can be used for rust disease detection are 680, 725, and 750 nm. Researchers have reported that more than 95% classification accuracy can be achieved using the Normalized Difference Vegetation Index (NDVI). Factors that cause stress in plants lead to disruption of the cell wall in the inner cavities of the leaves and reduced reflection in the near-infrared (NIR). Thus, these changes in the visible and NIR region can reveal changes in vegetative intensity, which can be used to detect plant diseases and insect damage (Hatfield & Pinter Jr, 1993; Nilsson, 1995).

Acknowledgments This study was carried out by Metin AYDOĞDU in the master's thesis "Determination of the Seasonal Effects of Different Iron and Zinc Applications on Yellow Rust (*Puccinia striiformis* f. sp. *tritici*) Disease in Winter Wheat Using Multi-Band Data" at Kırşehir Ahi Evran University, Institute of Science, Department of Agricultural Biotechnology. (YÖK Thesis No: 671046/Date: 25.05.2021). We would like to express our gratitude to the Department of Plant Diseases at the Field Crops Central Research Institute for their invaluable assistance in the preparation and execution of this thesis. Our sincere thanks also go to Dr. Nilüfer AKCI and the members of the thesis jury: Prof. Dr. Hikmet GÜNAL from the Department of Soil Science and Plant Nutrition at Harran University, and Dr. Nurullah ACİR from the same department at Kırşehir Ahi Evran University, for their significant contributions."

Funding Open access funding provided by the Scientific and Technological Research Council of Türkiye (TÜBİTAK).

Declaration

Competing interests There is no conflict of interest between authors.

Credit authorship contribution statement The authors declare their contributions to the manuscript such as the following sections:

Metin Aydoğdu: Investigation, Methodology Writing, Kadir AKAN: Investigation, Review of relevant literature, review, and editing.

Ethics committee decision The article does not require any ethical committee decision.

Appendix

Symbol	Detail	Symbol	Detail
NDVI	Normalized Difference Vegetation Index	CI	Coefficient infection
DI %	Disease Indice	DS %	Disease Severity
RMSE	Root Mean Square Error	SMA	Spectral Mixture Analysis
R ²	Pearson Correlation Value	NIR	Near-infrared
YRI	Yellow Rust Indice	SVI	Spectral vegetation indices
SE	Standard error	RE	Relative error
RGB	Red Green Blue	LC	Little Clup
Fe	Iron	Zn	Zinc

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