Spectral Reflectance Indices as a Potential Indirect Selection Criteria for Wheat Yield under Irrigation

M. A. Babar, M. P. Reynolds, M. van Ginkel, A. R. Klatt,* W. R. Raun, and M. L. Stone

ABSTRACT

The objectives of this study were to assess the potential of using spectral reflectance indices (SRI) as an indirect selection tool to differentiate spring wheat (*Triticum aestivum* L.) genotypes for grain yield under irrigated conditions. This paper demonstrates only the first step in using the SRI as indirect selection criteria by reporting genetic variation for SRI among genotypes, the effect of phenology and year on SRI and their interaction with genotypes, and the correlations between SRI and grain yield and yield components of wheat. Three field experiments—15 CIMMYT globally adapted genotypes (GHIST), 25 random F_3–derived lines (RLs1), and 36 random F_3–derived lines (RLs2)—were conducted under irrigated conditions at the CIMMYT research station in northwest Mexico in three different years. Five previously developed SRI (photochemical reflectance index [PRI], water index [WI], red normalized difference vegetation index [RNDVI], green normalized difference vegetation index [GNDVI], simple ratio [SR]) and two newly calculated SRI (normalized water index-1 [NWI-1] and normalized water index-2 [NWI-2]) were evaluated in the experiments. In general, genotypic variation for all the indices was significant. Near infrared radiation (NIR)–based indices (WI, NWI-1, NWI-2) gave the highest levels of association with grain yield during the 3 yr of the study. A clear trend for higher association between grain yield and the NIR-based indices was observed at heading and grainfilling than at booting. Overall, NIR-based indices were more consistent and differentiated grain yield more effectively compared to the other indices. The results demonstrated the potential of using SRI as a tool in breeding programs for selecting for increased genetic gains for yield.

SINCE THE GENETIC BASIS of yield improvement in wheat is not well established (Reynolds et al., 1999), the classical breeding approach for yield improvement still relies on an informed "numbers game" where crosses are made among potentially complementary parents. Subsequently, large numbers of their progeny have to be assessed visually in early generations and in yield trials as advanced lines to identify suitable materials to test in the target environment (Jackson, 2001). Classical breeding programs consider grain yield per se as the main selection criterion for grain yield (Loss and Siddique, 1994), but due to the high genotype × environment interaction component of this trait, commonly used statistical procedures are frequently not powerful enough to accurately differentiate between genotypes (Bhatti et al., 1991), thereby increasing the risk of accidentally discarding good lines or retaining inappropriate genotypes in trials (Ball and Konzak, 1993). Nonetheless, field evaluations are still necessary to effectively identify superior genotypes in a real-life setting, which is expensive in terms of time and financial resources especially when a large number of genotypes are being evaluated. Moreover, often additional evaluations are necessary in successive years and in different locations. To avoid or at least to reduce this laborious, time-consuming, and cumbersome process, an easy, rapid, and inexpensive selection tool may help breeders reliably screen large numbers of genotypes in a relatively short time before initiating expensive yield trials (Reynolds et al., 1999). It would be very advantageous if such a selection tool has higher heritability than grain yield, shows a strong correlation with grain yield, and could detect high yielding genotypes rapidly and efficiently from a large number of genotypes.

The use of morphological and physiological selection criteria to differentiate grain yield is an indirect breeding approach. Physiological tools have had limited utility in plant breeding programs (Jackson et al., 1996), partly because of the time-consuming evaluation methods and the lack of association with yield (Loss and Siddique, 1994; Richards, 1996). Nonetheless, canopy temperature, which can be sensed remotely using infrared thermometry, has been shown to be well associated with the yield of wheat cultivars (Reynolds et al., 1994; Fischer et al., 1998), as well with the yield of recombinant inbred lines and advanced breeding materials (Reynolds et al., 1998; 1999) in irrigated, high radiation environments.

More recent studies suggest that spectral reflectance is another promising remote sensing technique for screening genotypes (Araus, 1996; Araus et al., 2001). Canopy light reflectance properties mainly based on the absorption of light at a specific wavelength are associated with specific plant characteristics. The spectral reflectance in the visible (VIS) wavelengths (400–700 nm) depends on the absorption of light by leaf chlorophyll and associated pigments such as carotenoids and anthocyanins. The reflectance in the VIS is low because of the high absorption of light energy by pigments. The reflectance

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Abbreviations: GHIST, global historic trials; GNDVI, green normalized difference vegetation index; LAI, leaf area index; NIR, near infrared radiation; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RLs1, random lines-1; RLs2, random lines-2; RNDVI, red normalized difference vegetation index; SR, simple ratio; SRI, spectral reflectance indices; VIS, visible wavelength; WI, water index.

of the NIR wavelengths (700–1300 nm) is high because of the multiple scattering of light by different leaf tissues (Knipling, 1970). Spectral reflectance indices were developed on the basis of simple mathematical formulae such as ratios or differences between the reflectance at given wavelengths (Araus et al., 2001). Simple ratio (SR = NIR/VIS) and normalized difference vegetation index [NDVI = (NIR − VIS)/(NIR + VIS)] were the first SRI developed, combining information from the VIS and NIR wavelengths. These indices were used to predict different vegetation parameters, such as green biomass and green leaf area index (LAI) (Tucker and Sellers, 1986). Spectral reflectance indices have also been developed based only on VIS, including the photochemical reflectance index [PRI = $(R_{531} - R_{570})/(R_{531} + R_{570})$] used to assess radiation use efficiency by the plants (Peñuelas et al., 1995), and also only on NIR, such as the water index (WI = R_{900}/R_{970}) to assess water status of the canopy (Peñuelas et al., 1993). Spectral reflectance indices have been widely reported by different authors to assess different physiological conditions of the canopy such as total dry matter, LAI, photosynthetic capacity (Sellers, 1987), as well as green LAI and fraction of photosynthetically active radiation absorption (Wiegand and Richardson, 1990; Baret and Guyot, 1991; Wiegand et al., 1991). Spectral reflectance indices have also proven to be useful in the assessment of early biomass and vigor of different wheat genotypes (Elliott and Regan, 1993; Bellairs et al., 1996), water status in gerbera (*Gerbera jamesonii*) and barley (*Hordeum vulgare* L.) (Peñuelas et al., 1993, 1997), and different pigment concentrations in the leaves of soybean [*Glycine max* (L.) Merr.] (Chappelle et al., 1992). The potential for using SRI to predict in-season grain yield have also been reported in wheat (Raun et al., 2001) and in maize (*Zea mays* L.) (Osborne et al., 2002) under water-stressed environments.

Attempts have been made to evaluate the potential use of SRI in plant breeding to differentiate genotypes for yield under well-watered and/or moisture-stressed conditions in wheat (Hatfield, 1981; Ball and Konzak, 1993; Aparicio et al., 2000; Royo et al., 2003) and soybean (Ma et al., 2001). The studies under moisture stressed conditions showed the potential of using spectral indices under such conditions, but under well-watered conditions the association between yield and existing indices (NDVI and SR) were weak.

The goal of this study was to evaluate a broader range of SRI as potential screening tools in irrigated, high yielding environments. Specific objectives of the present study were to (i) evaluate the correlation of existing spectral indices with yield and agronomic traits of bread wheat genotypes under near optimum nitrogen and irrigation levels, (ii) derive new improved SRI that distinguish among high yielding genotypes better than pre-existing indices, and (iii) to determine the best growth stage to apply the spectral reflectance tool.

MATERIALS AND METHODS

Three experiments were conducted under irrigated conditions in three cropping seasons (years 2001–2002, 2002–2003, and 2003–2004) at the CIMMYT (International Maize and Wheat Improvement Center) experimental station near Ciudad Obregon in Sonora, Mexico (27°33′ N, 109°09′ W, 38 m above sea level). The soil type at the experimental station is a coarse sandy clay, mixed montmorillonitic type caliciorthid, low in organic matter and slightly alkaline (pH 7.7) in nature (Sayre et al., 1997). The weather is mostly sunny and dry during the winter cropping cycle. The experiments were planted in a bed planting system where each 5-m-long plot consisted of two beds and the distance between bed centers was 80 cm. Plot area was 8.0 m^2 (1.6 by 5 m). In the first 2 yr, three rows were planted on each bed with 15-cm distance between rows. In the third year, two rows were planted on the beds with a 20-cm inter-row spacing.

The seeding rate for each experiment was 78 kg ha^{-1} and the experiments were planted in the last week of November. Nitrogen and phosphorus were applied to the plots at rates of 200 kg ha^{-1} and 26 kg ha^{-1}, respectively. During the first 2 yr, 150 kg N and all of the P were applied during land preparation, and 50 kg N was applied in the second week of January coinciding with the first node growth stage and the second supplementary irrigation. In the third year, the same procedure was followed, but N was supplied at half the dosage during planting and half with the second supplementary irrigation. A total of five supplementary irrigations were applied in the first and third years, but in the second year only four supplementary irrigations were given. Folicur 250EW (25% tebuconazole) was applied twice in every crop cycle, in the second week of February (early booting) and in the second week of March (just after flowering) at the rate of 0.5 L ha^{-1} to protect the experimental materials from prevalent leaf rust (caused by *Puccinia triticina* Eriks).

Harvested area for grain yield was 4.8 m^2 (1.6 by 3 m). Before harvesting, 100 tillers with spikes were cut at the ground level to estimate the various yield components. The collected 100 tillers were oven dried at 75°C for 48 h. The weight of the oven-dried 100 tillers was measured, and then the tillers were threshed to calculate the harvest index. Two hundred grains were randomly collected from the harvested plots to estimate 1000-grain weight. Harvested grain yield was converted to grain yield in megagrams per hectare (Mg ha^{-1}). From harvest index, grain weight of 100 tillers, 1000-grain weight, and grain yield per unit area, other yield components, including spikes per square meter, grains per spike, and biomass at maturity were calculated.

Experimental Materials

Experiment 1: In the first experiment, we used 15 worldwide-adapted spring bread wheat genotypes developed by the wheat-breeding program of CIMMYT. The genotypes represent the historical success achieved by the breeding program at CIMMYT. While all are high yielding in distinct regions around the world, they vary widely in morphological traits and in parentage. The genotypes were planted in a 5 by 3 α-lattice design with two replications. In this paper we will refer to this experiment as "GHIST," since it studies a global historical set of commercial genotypes.

Experiment 2: This experiment had 25 genotypes, comprising 23 random F_3–derived lines and the two parents Sonalika and Attila. Approximately 1000 F_2 seeds were planted and the population was harvested in bulk. Next year approximately 500 F_3 seeds were space planted and random individual F_3 plants were selected. The selected F_3 plants were harvested separately and $F_{3:4}$ families were planted in separate small plots. The $F_{3:4}$ small plots were harvested separately (F_5 seed) and planted in individual $F_{3:5}$ yield plots that were harvested separately. The process was continued for two more generations to produce $F_{3:7}$ families. The experiment was planted in a 5 by 5 α-lattice design with two replications. In this paper we will refer to this experiment as "RLs1."

Experiment 3: We used 36 genotypes, comprising 34 random F_3–derived lines and their two parents (Bacanora 88 and Cndo/R143//Ente/Mexi_2/3/Ae. sq.(Taus)/4/Weaver). The random lines were developed in the same procedure as described for RLs1. The experiment was planted in a 6 by 6 α-lattice design with two replications. In this paper we will refer to this experiment as "RLs2."

Radiometric Measurements

The spectral reflectance measurements were taken by a portable narrow-bandwidth Spectroradiometer (Model Field-Spec UV/VNIR, Analytical Spectral Devices, Boulder, CO) with a 25° field of view. This instrument can detect reflected light from the canopy ranging from 350 to 1100 nm. Therefore, it covers VIS and NIR. It gives 512 continuous bands with a sampling interval of 1.43 nm. The spectroradiometer was connected to a computer, which stored the individual scans for subsequent processing. Each reflectance measurement was the average of 10 scans (which was programmed and calculated by the software used to operate the spectroradiometer) and the scanning area was approximately 18.94 cm^2. The sensor was mounted with the help of a pistol grip 40 to 50 cm above the canopy facing the center of the bed. The spectroradiometer was recalibrated against a white reference plate ($BaSO_4$) every 10 plots. The reflectance measurements were taken between 1030 to 1400 h under sunny conditions, and reflectance measurements were taken from four different places within each plot. The mean of the four readings was used to calculate spectral indices of each individual plot for statistical analysis. The average time required for the completion of the reflectance measurement from four different areas within the plot was approximately 40 to 45 s $plot^{-1}$.

The spectral reflectance measurements were taken at booting (Zadoks' stage between 39 and 47), heading (Zadoks' stage between 55 and 69), and grain filling (Zadoks' stage between 75 and 83) in all experiments (Zadoks et al., 1974), except for RLs1 and RLs2 in the year 2001–2002, where the reflectance measurements were taken only at the heading stage.

Calculation and Selection of Indices

Initially, different ratios and normalized indices were calculated based on a combination of visible and near-infrared wavelengths. From the combinations tested, two indices were selected for presentation in this paper. Those two indices combined information from 850, 900, and 970 nm. The 970 nm has been reported as a weak water absorption band (Peñuelas et al., 1993), and the other two bands (850 and 900 nm) were used as reference bands. We have referred to these two indices as normalized water index-1 (NWI-1) and normalized water index-2 (NWI-2). Five other reference indices, including the most widely used NDVI and SR, were calculated (described below) and compared with the two new indices.

The notation R_i was used to indicate the reflectance of light at a wavelength of *i* nm. The different SRI calculated were: PRI = $(R_{531} - R_{570})/(R_{531} + R_{570})$ (Peñuelas et al., 1995), which is an indicator of radiation use efficiency by the plants; WI = R_{970}/R_{900} (Peñuelas et al., 1993), which indicates canopy water status; RNDVI = $(R_{780} - R_{670})/(R_{780} + R_{670})$ (Raun et al., 2001), which indicates canopy photosynthetic area; GNDVI = $(R_{780} - R_{550})/(R_{780} + R_{550})$ (Gitelson et al., 1996), which indicates canopy photosynthetic area; SR = R_{900}/R_{680} (Aparicio et al., 2000), which is also an indicator of canopy photosynthetic active area. The above mentioned normalized water indices were calculated as follows: NWI-1 = $(R_{970} - R_{900})/(R_{970} + R_{900})$, and NWI-2 = $(R_{970} - R_{850})/(R_{970} + R_{850})$.

Statistical Analysis

Alpha-lattice analyses for grain yield and spectral indices were performed using PROC MIXED procedure of the SAS/STAT statistical package (SAS Institute, 2001). Data were analyzed in each individual growth stage within the same year and between the years. Combined analyses were performed across different growth stages and different years (years 2002–2003 and 2003–2004 for RLs1 and RLs2, and all 3 yr for GHIST). Mean squares of combined analysis were obtained by using PROC MIXED following type1 method. Pearson correlation coefficients were used to estimate the relationships of yield and yield components with different spectral indices and the relationships of indices at different growth stages within the same year and between years. Genetic correlations between traits were estimated using PROC MIXED, using a program following the method described by Singh and Chaudhary (1977).

RESULTS

Genotypic Performance

Minimum, maximum, mean, LSD, and significance level of *F*-test within and across years for grain yield in three different experiments are presented in Table 1. The genotypic variations for grain yield in all three experiments within and across years were significant. The minimum and maximum mean SRI values over different growth stages and years in the GHIST experiment were −0.054 to −0.027 (PRI), 0.838 to 0.875 (WI), 0.820 to 0.893 (RNDVI), 0.723 to 0.893 (GNDVI), 14.2 to 26.6 (SR), −0.088 to −0.067 (NWI-1), and −0.084 to −0.059 (NWI-2). The SRI values in the RLs1 experiment ranged from −0.055 to −0.032 (PRI), 0.862 to 0.887 (WI), 0.854 to 0.904 (RNDVI), 0.754 to 0.798 (GNDVI), 12.3 to 19.3 (SR), −0.074 to −0.060 (NWI-1), and −0.069 to −0.052 (NWI-2). For RLs2, the range of SRI was −0.0594 to

Table 1. Minima, maxima, means, and LSDs for pairwise genotypic comparisons and significance level of grain yield (Mg ha^{-1}) within and between years in three different experiments.

	2001–2002	2002–2003	2003–2004	Across years
		GHIST		
Minimum	5.23	4.91	5.28	4.97
Maximum	7.32	7.40	7.07	7.50
Mean	6.49	5.92	5.86	6.09
LSD(5%)	0.51	0.96	0.58	0.73
Significance level	*	*	**	**
		RLs1		
Minimum	4.16	4.17	3.68	3.62
Maximum	6.49	6.47	5.41	6.47
Mean	5.43	5.50	4.56	5.16
LSD(5%)	0.54	1.00	0.56	0.72
Significance level	**	*	**	**
		RLs2		
Minimum	4.90	5.00	4.16	4.15
Maximum	7.91	7.26	5.92	7.91
Mean	6.37	6.23	5.29	5.96
LSD(5%)	0.73	0.93	0.80	0.86
Significance level	**	**	**	**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

−0.0333 (PRI), 0.836 to 0.874 (WI), 0.821 to 0.900 (RNDVI), 0.741 to 0.807 (GNDVI), 14.3 to 23.8 (SR), −0.0893 to −0.0671 (NWI-1), and −0.0856 to −0.0610 (NWI-2). Genotypes showed significant variation for SRI in all three experiments (data not presented).

Effect of Growth Stages

Minimum, maximum, mean, LSD, and significance level for these SRI at the three different growth stages (booting, heading, and grainfilling) are presented in Table 2. The analysis of spectral indices at individual growth stages across the years revealed significant genotypic variation for the indices at all growth stages. In general, there was less variation among genotypes for the spectral indices at the booting stage than at the later growth stages. This was evidenced by the fact that when considering individual years, not all indices showed significant differences among genotypes when measured at booting (data not shown).

Regarding the main effect of growth stages on SRI, the values based on NIR (WI, NWI-1, and NWI-2) tended to decrease from booting to heading, and then increased in the grainfilling stage (Table 2, Fig. 1). The only index based on visible wavelength, PRI, showed the highest value at the booting stage, with values decreasing as the growth cycle progressed (Table 2). The values of RNDVI, GNDVI, and SR (based on red, green, and near infrared radiation) were similar at the booting and heading stages, while their values decreased during grainfilling.

Genotype, Growth Stage, and Year Interactions

Mean squares from the analysis of variance combined over growth stages and years for the GHIST experiment are presented in Table 3. The genotypic main effect in the combined analysis was significant for all indices and grain yield. Genotypes × growth stages and genotype × year interactions were significant for all the indices except for RNDVI. The genotype × year interaction for grain yield was also highly significant. Similar trends for genotypic and interactions effects were also observed in two other experiments for different SRI (data not shown). The associations of each SRI measured in different growth stages within the same year, and between the years are presented for the experiment GHIST in Tables 4 and 5. In general, a strong association was ob-

Table 2. Minima, maxima, means, and LSDs for pairwise genotypic comparisons and significance level of spectral indices at individual growth stages across years.†

		GHIST‡			RLs1§			RLs2§		
		Boot	Hd	GF	Boot	Hd	GF	Boot	Hd	GF
PRI	**Minimum**	−0.039	−0.058	−0.105	−0.045	−0.055	−0.119	−0.039	−0.046	−0.117
	Maximum	−0.005	−0.015	−0.035	−0.007	−0.013	−0.036	−0.008	−0.017	−0.046
	Mean	−0.023	−0.028	−0.056	−0.024	−0.031	−0.088	−0.024	−0.029	−0.083
	LSD(5%)	0.004	0.005	0.008	0.008	0.015	0.014	0.004	0.007	0.013
	Significance level	*	**	**	*	**	**	*	**	**
WI	**Minimum**	0.826	0.774	0.831	0.838	0.795	0.832	0.865	0.819	0.853
	Maximum	0.899	0.885	0.900	0.886	0.881	0.915	0.908	0.883	0.921
	Mean	0.876	0.832	0.857	0.861	0.834	0.875	0.889	0.852	0.886
	LSD(5%)	0.027	0.024	0.022	0.024	0.026	0.023	0.026	0.021	0.025
	Significance level	*	**	**	*	**	**	*	**	**
RNDVI	**Minimum**	0.810	0.793	0.684	0.856	0.861	0.639	0.902	0.878	0.549
	Maximum	0.953	0.954	0.891	0.938	0.928	0.922	0.948	0.941	0.864
	Mean	0.900	0.894	0.834	0.903	0.902	0.823	0.927	0.917	0.742
	LSD(5%)	0.031	0.052	0.035	0.024	0.036	0.050	0.011	0.011	0.076
	Significance level	*	**	**	*	**	**	*	**	**
GNDVI	**Minimum**	0.712	0.673	0.619	0.723	0.753	0.625	0.781	0.769	0.554
	Maximum	0.869	0.874	0.793	0.837	0.833	0.824	0.851	0.847	0.767
	Mean	0.792	0.790	0.736	0.780	0.797	0.734	0.816	0.817	0.679
	LSD(5%)	0.029	0.071	0.032	0.028	0.043	0.046	0.019	0.018	0.052
	Significance level	*	**	**	*	**	**	*	**	**
SR	**Minimum**	15.9	10.0	5.6	12.7	14.1	3.1	18.9	15.0	3.5
	Maximum	40.3	40.2	17.0	30.7	26.3	13.8	37.2	31.9	13.3
	Mean	25.7	22.9	11.8	20.1	19.9	6.7	26.2	23.2	7.3
	LSD(5%)	3.55	5.66	2.12	3.92	5.83	1.91	4.02	3.31	2.23
	Significance level	**	**	**	*	**	**	*	**	**
NWI-1	**Minimum**	−0.097	−0.128	−0.093	−0.071	−0.100	−0.080	−0.088	−0.114	−0.091
	Maximum	−0.052	−0.059	−0.051	−0.048	−0.062	−0.040	−0.061	−0.064	−0.043
	Mean	−0.066	−0.092	−0.077	−0.059	−0.080	−0.060	−0.075	−0.090	−0.066
	LSD(5%)	0.012	0.009	0.009	0.011	0.009	0.011	0.011	0.011	0.009
	Significance level	*	**	**	*	**	**	**	**	**
NWI-2	**Minimum**	−0.094	−0.127	−0.084	−0.068	−0.096	−0.073	−0.084	−0.112	−0.082
	Maximum	−0.043	−0.052	−0.041	−0.045	−0.056	−0.023	−0.057	−0.065	−0.028
	Mean	−0.060	−0.087	−0.069	−0.055	−0.074	−0.049	−0.071	−0.089	−0.055
	LSD(5%)	0.012	0.009	0.011	0.012	0.011	0.014	0.011	0.011	0.012
	Significance level	*	**	**	*	**	**	*	**	**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† Boot, booting stage; GF, grainfilling stage; GHIST, global historic trials; GNDVI, green normalized difference vegetation index; Hd, heading stage; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RLs1, random lines-1; RLs2, random lines-2; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

‡ Estimates were calculated based on 3 yr (2001–2002, 2002–2003, and 2003–2004) in GHIST.

§ Estimates were calculated based on 2 yr (2002–2003 and 2003–2004) in RLs1 and RLs2.

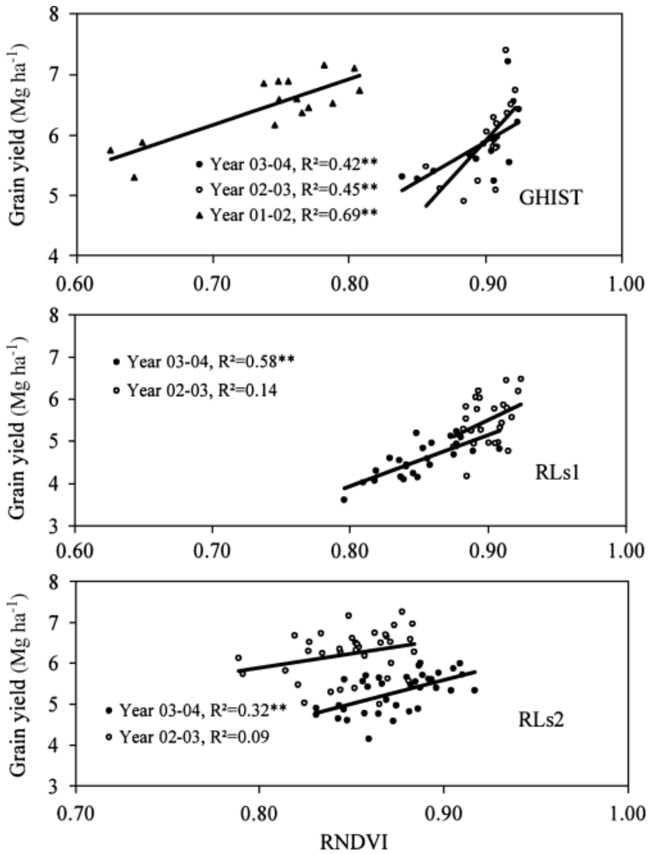

Fig. 1. The relationship between grain yield and red normalized difference vegetation index (RNDVI) in three different experiments in different years where grain yields of the genotypes were plotted against the mean values of the index averaged over three growth stages within the years. **Significant at 0.01 probability level.

served for all indices measured at heading and grainfilling in all 3 yr (Table 4). The indices RNDVI, GNDVI, and SR also showed a strong association between booting and heading, and booting and grainfilling in the years 2002–2003 and 2003–2004, but not in the year 2001–2002. The associations between booting and heading, and booting and grainfilling for the three NIR-based indices (WI, NWI-1, and NWI-2) and for PRI were very low in all 3 yr.

All indices showed the highest association between years at the grainfilling stage except for PRI and GNDVI between years 2001–2002 and 2002–2003 (Table 5). RNDVI, GNDVI, and SR also showed a strong association between years 2002–2003 and 2003–2004 at the booting stage, but not between years 2001–2002 and 2002–2003. The NIR-based indices showed low between-year correlations for that growth stage.

Association of Grain Yield with SRI

The correlation of grain yield with spectral indices at (i) different growth stages for each year, (ii) averaged across growth stages for each year (mean), and (iii) averaged over growth stages and years (overall mean) are presented in Table 6. The association between mean SRI (averaged over growth stages within an individual year) and the overall grain yield (mean of 3 yr) are presented in Table 7. We calculated 72 different ratios and normalized indices by using different visible and NIR wavelength combinations and also calculated 20 previously published SRI which were indicative of different physiological conditions of plants. Initially, we selected five out of 72 SRI that were calculated but later selected only two of them to present in this paper because of their high correlation with grain yield and consistent performance over years and different genetic backgrounds. Of the previously published SRI, we selected five to present in this paper. RNDVI, GNDVI, and SR have been the most widely used by different authors to study the physiological conditions of plants, and all five SRI (RNDVI, GNDVI, SR, WI and PRI) have been reported by various authors to differentiate genotypes for grain yield under water-stressed conditions in durum wheat (*Triticum turgidum* L. subsp. *durum*) and well-watered conditions in bread wheat and soybean.

Comparing all the indices, those based on NIR (WI, NWI-1, and NWI-2) demonstrated a higher level of association with grain yield compared with the other spectral indices (RNDVI, GNDVI, SR, and PRI) at the heading and grainfilling stages, except for GHIST in

Table 3. Mean squares of the combined analysis of variance across different growth stages and years for different spectral reflectance indices and across different years for grain yield in GHIST.†

Effect	DF‡	PRI	WI	RNDVI	GNDVI	SR	NWI-1	NWI-2	Yield
Year	2 (2)	0.00406**	0.013029**	0.139385**	0.173776*	1645.4**	0.005682**	0.006128**	0.1524*
Rep (year)	3 (3)	0.000015	0.000212	0.005233	0.010159	10.98	0.000085	0.000048	0.0104
Subk (year × rep)	24 (24)	0.000040	0.000214	0.000383	0.000282	10.35	0.000071	0.000084	0.0419
Entry	14 (14)	0.001262**	0.001699**	0.007741**	0.009867**	166.14**	0.000587**	0.000676**	0.2132**
Year × entry	28 (28)	0.00004**	0.00018*	0.000685	0.000667*	10.14*	0.000048*	0.000048*	0.1738**
Rep × entry (year)	18	0.000005	0.000083	0.000301	0.000309	4.20	0.00002	0.000020	
GS	2	0.027485**	0.041755**	0.119478**	0.08999**	4871.8**	0.014882**	0.017337**	
Year × GS	4	0.000255**	0.000427**	0.002451**	0.001787**	17.50**	0.000142**	0.000169**	
GS × entry	28	0.000902**	0.008644**	0.024987**	0.028141**	662.46**	0.003116**	0.003316**	
Year × GS × entry	56	0.000033**	0.000149**	0.00043	0.000307	6.80*	0.000049**	0.000064**	
Residual	90 (18)	0.000007	0.000081	0.000623	0.000893	4.14	0.000028	0.000035	0.0571
Total	269 (89)								

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† GNDVI, green normalized difference vegetation index; GS, growth stages; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

‡ Numbers in the parenthesis indicate the DF of respective source of variation for grain yield.

Table 4. The relationship of spectral indices among different growth stages within years in GHIST.†

	PRI		WI		RNDVI		GNDVI		SR		NWI-1		NWI-2	
	Hd	GF	Hd	GF	Hd	GF	Hd	GF	Hd	GF	Hd	GF	Hd	GF
2001–2002														
Boot	0.451	0.500	0.290	0.059	0.178	0.145	0.404	0.144	0.548*	0.331	0.331	0.086	0.287	0.073
Hd		0.884**		0.886**		0.919**		0.896**		0.910**		0.885**		0.848**
2002–2003														
Boot	0.481	0.501	0.094	0.204	0.677**	0.723**	0.792**	0.759**	0.599**	0.613*	0.097	0.257	0.149	0.318
Hd		0.869**		0.729**		0.716**		0.686**		0.780**		0.715**		0.731**
2003–2004														
Boot	0.196	0.124	0.197	0.024	0.898**	0.771**	0.844**	0.616**	0.874**	0.715**	0.236	0.037	0.060	0.050
Hd		0.943**		0.922**		0.943**		0.919**		0.915**		0.933**		0.932**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† Boot, booting stage; GF, grainfilling stage; GHIST, global historic trials; GNDVI, green normalized difference vegetation index; Hd, heading stage; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

year 2001–2002. Near infrared radiation–based indices showed significant negative correlations with grain yield at heading and grainfilling stages in all the experiments in all 3 yr. The other indices (i.e., PRI, RNDVI, GNDVI, SR) generally showed significant positive relationships with grain yield. Of all the indices calculated, PRI showed the least predictive capacity to differentiate genotypes for grain yield. When considering indices averaged over growth stages, NIR-based indices gave the highest correlations with grain yield except for GHIST in year 2001–2002.

At booting, RNDVI, GNDVI, SR. and PRI were better correlated with grain yield than the NIR-based indices. Near infrared radiation–based indices showed a very low level of association with grain yield at the booting stage, which was consistent across all the experiments and in all the years. In most cases, mean indices over three growth stages correlated better with grain yield than any single growth stage with a few exceptions. A similar trend has also been observed when the mean indices over two growth stages (heading and flowering) were used, and the correlation values were very close to the correlation values when the mean indices over three growth stages were used, with a few exceptions (data not shown). The correlations between the overall mean indices (across different growth stages and years) showed very strong correlations with the overall mean grain yield of genotypes (across different years) and the correlations values were higher than between SRI and grain yield within an individual year.

The three NIR-based indices showed a very strong association with overall mean grain yield (mean of 3 yr), when the mean indices over three growth stages within an individual year were correlated with overall mean grain yield. The levels of association were consistently higher than the association between other SRI and overall mean grain yield with a single exception (Table 7).

No single NIR-based index showed any definite superiority over the others. Nonetheless, when we consider all the correlations between these three indices and grain yield within year, between indices measured in an individual year and overall mean grain yield, and between overall mean indices and overall mean grain yield, NWI-2 gave either similar or marginally better correlations than the other two NIR-based indices (WI and NWI-1). For simplicity of the presentation in the regression model, we have used only NWI-2.

Attempts were made to determine the most suitable regression model to explain variability among the genotypes for grain yield across different years (Fig. 1,2). NWI-2 and RNDVI were compared in different regression models because of the performance of NWI-2, and because the RNDVI has been the most widely used SRI to study grain yield variations at the genotypic level, as well as in agronomic trials. In general, a simple linear model did not differ greatly from exponential and power regression models in explaining grain yield variation (except in GHIST). The exponential model explained 2 to 3% more of the variability for grain yields for the different indices in year 2003–2004. A simple linear model was equally applicable in explaining the phenological pattern of the relationship between grain yield and the indices (except for GHIST) (data not presented). The exponential model explained 2 to 3% more of the

Table 5. The relationship of spectral indices between years at different growth stages in GHIST.†

	PRI	WI	RNDVI	GNDVI	SR	NWI-1	NWI-2
Between 2001–2002 and 2002–2003							
Booting	0.508	0.305	0.326	0.465	0.614*	0.306	0.301
Heading	0.855**	0.726**	0.848**	0.873**	0.823**	0.722**	0.703**
Grainfilling	0.831**	0.803**	0.852**	0.816**	0.844**	0.767**	0.792**
Between 2002–2003 and 2003–2004							
Booting	0.507	0.581*	0.829**	0.825**	0.771**	0.571*	0.519*
Heading	0.863**	0.624*	0.731**	0.710**	0.771**	0.628*	0.658**
Grainfilling	0.904**	0.855**	0.885**	0.912**	0.915**	0.848**	0.848**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† GNDVI, green normalized difference vegetation index; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

Table 6. Correlation coefficients between grain yield and different spectral indices at different growth stages in different years.†

	2001–2002				2002–2003				2003–2004				
Indices	Boot	Hd	GF	Mean‡	Boot	Hd	GF	Mean‡	Boot	Hd	GF	Mean‡	Overall mean§
							GHIST						
PRI	0.580*	0.692**	0.785**	0.691**	0.451	0.662**	0.635*	0.721**	0.648**	0.645**	0.637*	0.709**	0.818**
WI	−0.139	−0.731**	−0.731**	−0.707**	−0.222	−0.719**	−0.739**	−0.740**	−0.438	−0.662**	−0.628*	−0.753**	−0.876**
RNDVI	0.518*	0.585*	0.707**	0.829**	0.750**	0.675**	0.590*	0.672**	0.639*	0.620*	0.595*	0.646**	0.787**
GNDVI	0.522*	0.751**	0.734**	0.814**	0.767**	0.747**	0.627*	0.753**	0.749**	0.646**	0.584*	0.698**	0.863**
SR	0.292	0.585*	0.692**	0.581*	0.825**	0.731**	0.628*	0.800**	0.629*	0.640**	0.643**	0.724**	0.812**
NWI-1	−0.136	−0.739**	−0.769**	−0.701**	−0.233	−0.758**	−0.740**	−0.751**	−0.464	−0.651**	−0.601*	−0.731**	−0.881**
NWI-2	−0.097	−0.676**	−0.709**	−0.703**	−0.092	−0.784**	−0.744**	−0.748**	−0.472	−0.687**	−0.708**	−0.802**	−0.872**
							RLs1						
PRI		0.188			0.216	0.058	−0.067	−0.003	0.455*	0.455*	0.591**	0.568**	0.354
WI		−0.549**			−0.305	0.651**	−0.418*	−0.718**	−0.272	−0.799**	−0.834**	−0.814**	−0.861**
RNDVI		−0.219			0.309	0.299	0.303	0.368	0.612**	0.648**	0.762**	0.759**	0.546**
GNDVI		−0.115			0.431*	0.399*	0.401*	0.478*	0.530**	0.624**	0.709**	0.722**	0.506**
SR		−0.235			0.354	0.396*	0.135	0.411*	0.553**	0.611**	0.659**	0.648**	0.408*
NWI-1		−0.588**			−0.343	−0.654**	−0.381	−0.748**	−0.229	−0.778**	−0.845**	−0.797**	−0.863**
NWI-2		−0.607**			−0.345	−0.605**	−0.308	−0.728**	−0.222	−0.786**	−0.828**	−0.803**	−0.870**
							RLs2						
PRI		0.262			0.507**	0.584**	0.279	0.456**	0.294	0.303	0.502**	0.479**	0.452**
WI		−0.578**			−0.554**	−0.605**	−0.569**	−0.667**	−0.419*	−0.716**	−0.723**	−0.773**	−0.743**
RNDVI		0.379*			0.111	0.434**	0.249	0.287	0.492**	0.557**	0.564**	0.598**	0.457**
GNDVI		0.551**			0.288	0.622**	0.250	0.408*	0.435**	0.579**	0.554**	0.589**	0.571**
SR		0.374*			0.119	0.439**	0.220	0.308	0.442**	0.551**	0.597**	0.582**	0.468**
NWI-1		−0.593**			−0.539**	−0.611**	−0.562**	−0.660**	−0.385*	−0.711**	−0.726**	−0.772**	−0.747**
NWI-2		−0.599**			−0.588**	−0.682**	−0.578**	−0.706**	−0.429**	−0.704**	−0.715**	−0.773**	−0.780**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† Boot, booting stage; GF, grainfilling stage; GHIST, global historic trials; GNDVI, green normalized difference vegetation index; Hd, heading stage; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RLs1, random lines-1; RLs2, random lines-2; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

‡ Correlation between yield and mean spectral indices averaged over three different growth stages within the same year.

§ Correlation between mean yield and mean spectral indices averaged over years.

variability for RNDVI and NWI-2 when the readings were taken at heading and grainfilling stages in year 2003–2004. As the nonlinear model was not significantly better than the linear model, simple linear models are presented in Fig. 1 and 2.

We selected the 20% top yielding genotypes for grain yield based on NDVI and NWI-2 in two populations (RLs1 and RLs2). First, we ranked the genotypes based on grain yield per se, and then ranked the genotypes based on the two indices. Sixty to eighty percent and 57 to 86% of the top 20% highest yielding genotypes were selected based on NWI-2 in RLs1 and RLs2, respectively, in the three different years of study. NDVI selected 20 to 80% and 43 to 71% of the top 20% genotypes in RLs1 and RLs2 experiments, respectively, in the 3-yr period of study.

Association of SRI with Agronomic Traits

The associations between SRI and two agronomic traits, grains per square meter and biomass at maturity, averaged over the 3 yr are presented in Table 8. The correlations between SRI and additional agronomic traits such as harvest index, spikes per square meter, grains

Table 7. The correlations between different spectral reflectance indices (averaged over growth stages within each individual year) and grain yield (mean of 3 yr) in three different experiments under irrigated conditions, presented by year.†

Years	PRI	WI	RNDVI	GNDVI	SR	NWI-1	NWI-2
				GHIST			
2001–2002	0.750**	−0.825**	0.719**	0.702**	0.625**	−0.822**	−0.827**
2002–2003	0.809**	−0.893**	0.813**	0.869**	0.880**	−0.801**	−0.792**
2003–2004	0.827**	−0.833**	0.802**	0.857**	0.756**	−0.818**	−0.861**
				RLs1			
2001–2002‡	−0.189	−0.500*	−0.212	−0.124	−0.220	−0.535**	−0.573**
2002–2003	0.201	−0.706**	0.131	0.247	0.404*	−0.739**	−0.753**
2003–2004	0.456*	−0.797**	0.677**	0.682**	0.590**	−0.794**	−0.799**
				RLs2			
2001–2002‡	0.375	0.514**	0.447**	0.578**	0.415*	−0.549**	−0.563**
2002–2003	0.407*	−0.717**	0.395*	0.479**	0.418*	−0.716**	−0.761**
2003–2004	0.420*	−0.583**	0.377	0.463**	0.395*	−0.575**	−0.573**

* Significant at 0.05 probability level.

** Significant at 0.01 probability level.

† GHIST, global historic trials; GNDVI, green normalized difference vegetation index; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RLs1, random lines-1; RLs2, random lines-2; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

‡ Correlation between the different SRI measured only at heading stage and grain yield (mean of 3 yr) in two experiments in a particular year.

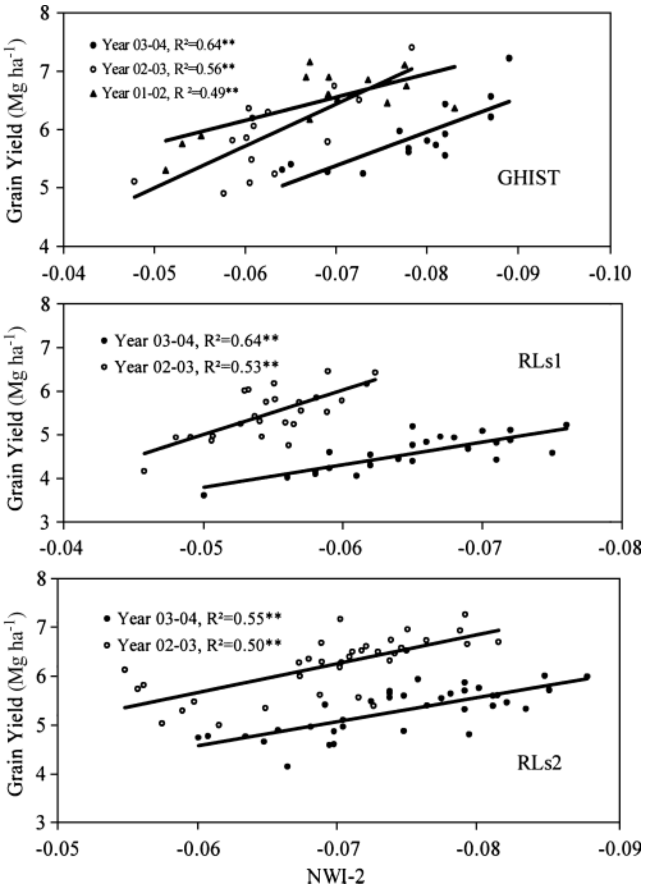

Fig. 2. Relationship between grain yield and normalized water index-2 (NWI-2) in three different experiments in different years where yields of the genotypes were plotted against mean value of the index of three different growth stages within the same year. ** Significant at 0.01 probability.

per spike, and 1000-grain weight were also evaluated. In general, the associations between SRI and these four agronomic parameters were low and inconsistent (not shown).

All indices showed significant association with grains per square meter and biomass at maturity in all three experiments. However, the three NIR-based indices were better correlated with these two parameters compared to the other indices in RLs1 and RLs2.

DISCUSSION

Genotypic Performance

In this study, we observed a wide range of genetic variation for different spectral indices, as was also shown in earlier studies under irrigated conditions (Ball and Konzak, 1993; Aparicio et al., 2000) and moisture-stressed conditions (Aparicio et al., 2000; Royo et al., 2003). For most indices, genotypes could not be distinguished from one another at the booting stage. This could be attributed to a more or less uniformly high LAI at booting, making it difficult for genotypes to be differentiated. Variation among genotypes increased at heading and during grainfilling, quite likely due to a decrease in LAI and differing morphological characteristics of the spikes. Asrar et al. (1984) and Ahlrichs and Bauer (1983) showed that NDVI and NIR reflectance were sensitive to changes in leaf area up to values of LAI 3, after which they reached a plateau. Calderini et al. (1997) reported that wheat under irrigated conditions achieved maximum LAI between the stages of terminal spikelet and booting (LAI > 5 for most of the genotypes), and then decreased toward anthesis. Hatfield (1981), in another study on wheat grown under irrigated conditions, observed only small differences among varieties in NDVI once 100% ground cover was reached, but variability among varieties again increased as the crop cycle progressed, and the highest variability was obtained at maximum head weight due to differences in spike size and/or morphology.

Effect of Growth Stages

In general, values of PRI, RNDVI, GNDVI, and SR decreased from heading to grainfilling (Table 2). Aparicio et al. (2000) also reported a similar trend of decreasing values for spectral indices (PRI, RNDVI, and SR) with the advancement of growth stages in durum wheat under irrigated conditions. As RNDVI, GNDVI, and SR are indices that combine red, green, and NIR wavelengths, a reduction in LAI would decrease the reflectance of NIR but increase reflectance of visible wavelengths. The overall result would cause a decrease in the values of these three indices at the grainfilling stage. This is consistent with the observation that LAI commonly peaks at the booting stage and decreases as the growth cycle progresses (Calderini et al., 1997; Aparicio et al., 2000).

Values for the NIR-based indices WI, NWI-1, and NWI-2 decreased from booting to heading, and then increased at grainfilling. These indices are based on reflectance at 970 nm, which is a weak water absorbance band (Peñuelas et al., 1993), and on reflectance at 900 nm and 850 nm, which is caused by multiple reflection and scattering of light in the spongy mesophyll structure (Knipling, 1970). The WI shows an inverse relationship with water status at both the canopy and leaf level (Peñuelas et al., 1993, 1997). Water index assesses the

Table 8. Relationship between spectral indices and grains per square meter and biomass at maturity (BM) averaged over 3 yr.†

	GHIST		RLs1		RLs2	
Indices	Grains m−²	BM	Grains m−²	BM	Grains m−²	BM
PRI	0.754**	0.408	0.433*	0.438*	0.308	0.503**
WI	−0.624*	−0.576*	−0.688**	−0.816**	−0.625**	−0.775**
RNDVI	0.749**	0.609*	0.624**	0.612**	0.423*	0.523**
GNDVI	0.759**	0.581*	0.562**	0.549**	0.478**	0.565**
SR	0.673**	0.659**	0.581**	0.580**	0.442**	0.477**
NWI-1	−0.633*	−0.582*	−0.704**	−0.820**	−0.628**	−0.781**
NWI-2	−0.573*	−0.587*	−0.727**	−0.847**	−0.626**	−0.821**

* Significant at 0.05 probability level.
** Significant at 0.01 probability level.
† GHIST, global historic trials; GNDVI, green normalized difference vegetation index; NWI-1, normalized water index-1; NWI-2, normalized water index-2; PRI, photochemical reflectance index; RLs1, random lines-1; RLs2, random lines-2; RNDVI, red normalized difference vegetation index; SR, simple ratio; WI, water index.

changes in relative water content, leaf water potential, stomatal conductance, and canopy temperature in plants (Peñuelas et al., 1993). Based on the values of WI and the other NIR-related indices, it appears that in general the plant canopy contains a higher total amount of water at heading compared to later growth stages. Assessment of fresh and dry biomass made at these growth stages on the above experiments indicated that there was 14 to 22% more water in the canopy at heading than at grainfilling (data not shown).

Genotype, Growth Stage, and Year Interactions

In this study, we observed significant interaction between growth stages and genotypes in regard to their index values (Table 3). The low association between booting and flowering, and booting and grainfilling for NIR-based indices and PRI (Table 4), indicates that the ranking of genotypes changed between growth stages. On the other hand, RNDVI, GNDVI, and SR showed a higher correlation between booting and flowering and between booting and grainfilling. The interactions of growth stages and indices indicate that care must be taken to identify a suitable growth stage at which the indices will be applied to discriminate most effectively among the genotypes in breeding trials. Aparicio et al. (2002) also reported a significant interaction between growth stages and spectral indices (NDVI and SR) in durum wheat.

Association of Grain Yield with SRI

Several authors (Ball and Konzak, 1993; Peñuelas et al., 1997; Aparicio et al., 2000; Ma et al., 2001; Royo et al., 2003) have reported on the potential use of different spectral indices (mostly in regard to NDVI and SR) to differentiate genotypes for grain yield under diverse environmental conditions. A large proportion of the variation in yield in barley under three salinity levels was explained by spectral indices (Peñuelas et al., 1997), in soybean with three different planting densities and two different soil types under well-watered conditions (Ma et al., 2001), and in durum wheat under different moisture levels and at different locations (Royo et al., 2003). A large part of the variation being explained in these studies may be driven by the diverse environmental conditions to which the crops were exposed. Royo et al. (2003) reported a very high variability in grain yield of different durum wheat genotypes when SRI were combined across environments in a stepwise regression model (nine different moisture levels, nine field experiments, and nine SRI were considered together), but when individual environments were studied on their own, the amount of variation explained decreased considerably. Their study did not give any clear indication of which SRI were suitable for use under different environmental conditions. However, Aparicio et al. (2000) did show a strong relationship between grain yield and spectral indices under rainfed conditions in durum wheat.

This study includes historical landmark genotypes developed by CIMMYT with high yield capacity and with considerable diversity in morphology, plus different sister lines (RLs) expressing a similarly large range of diversity. Strong, consistent phenotypic correlations were observed between the NIR-based indices and grain yield in all experiments in three successive years. Also, high genotypic correlations between grain yield and the NIR-based spectral indices at the heading and grainfilling stages were observed (genotypic correlation values ranged from -0.594 to -0.925 in the above mentioned three different experiments) (data not shown). The estimation of genotypic correlations was done on small genotypic sample sizes in our study and may not be very reliable, but their magnitude should reflect true population values. This indicates that when using the NIR-based indices, the high amount of variation explained is related to variability among genotypes, and not due to any large environmental effects.

The spectral indices based on NIR (WI, NWI-1, and NWI-2) generally showed negative correlations with yield, increasing in value with the advancement of the crop cycle in all experiments (Table 6). These indices all incorporate an indicator of water status in the canopy. With an increasing amount of water in the plant, a decreasing amount of energy at 970 nm is reflected. Hence, the negative sign of the correlations reflect the fact that an increasingly low water status is associated with decreasing yields. Similar results were reported by Peñuelas et al. (1997) and Royo et al. (2003), both under irrigated and water-stressed conditions in durum wheat and barley.

RNDVI, GNDVI, and SR gave significant positive correlations with grain yield at the heading and grainfilling stages in nearly all cases over the 3 yr. Ball and Konzak (1993) and Royo et al. (2003) reported a significant positive correlation between grain yield and RNDVI at grainfilling for spring and durum wheat under well-watered conditions. On the other hand, Aparicio et al. (2000) found a significant correlation between grain yield and RNDVI only at the maturity stage, and not at the other growth stages (booting, heading, anthesis, and milk-grain) in durum wheat under irrigated conditions. We have observed that in most cases RNDVI, GNDVI, and SR showed an increasingly higher correlation with grain yield as growth progressed from booting to flowering or grainfilling (Table 6). Also Ma et al. (2001), when studying soybean, found a clear increasing trend in the correlation values between NDVI and grain yield from full flowering (R2) to seed formation (R5) at three different planting densities in two soil types under well-watered conditions.

All indices studied explained the largest amount of the variation when taken at the heading or grainfilling stages. Our results generally agree with the findings of Ma et al. (2001) in soybean and Royo et al. (2003) in durum wheat under well-watered conditions. In our study it is evident that the mean values over different growth stages gave higher correlations with grain yield, which was not reported by previous authors. The three NIR-based indices measure the water status at the canopy level, while RNDVI, GNDVI, and SR measure the greenness of the canopy, and PRI is an indicator of radiation use efficiency in plants. All the indices are indicative of healthy plant conditions in the field. The repeated measurements on the same genotypes at different growth stages basically accumulate information on the health or condition of the genotypes over a period of time. We hypothesize that the mean values of the indices for the different growth stages represent cumulative information on the health of the canopy, which translates into a higher correlation with final grain yield. This may also be an indication that the underlying genetic correlation is stronger than the phenotypic one.

Since the time required to take the SRI data in the field is just 40 to 45 s plot^{-1}, it would be desirable to take the readings more than once. At a minimum, one measurement at heading and another one at grainfilling might effectively differentiate genotypes for grain yield. Hence, those two stages appear to be the most appropriate time to apply these spectral indices if the objective is to discriminate genotypes for grain yield. Very strong correlations were observed between the overall mean of the SRI and overall grain yield. This strong correlation is also evidence of improved association between SRI and grain yield when more dates were averaged over years and growth stages, which was not reported before.

Our study also demonstrated a high efficiency of the SRI to evaluate the yield performance of genotypes over a period of time, which might be one of the most critical questions for breeders in evaluating a genotype for a particular environment (Table 7). The three NIR-based indices were more successful than others. Our study also showed a very high efficiency for SRI to select superior genotypes for grain yield, and NWI-2 performed better than RNDVI in selecting the top yielding genotypes for grain yield. These results are a definitive indication of the efficiency of NIR-based SRI for selecting superior genotypes for grain yield production.

The performance of the three NIR-based indices was very similar in explaining grain yield variability among genotypes within a year or over a period of time. Normalizing the water index (NWI-1 and NWI-2) did not significantly improve the relationship. Tucker (1979) showed the superiority of a normalized index over a ratio index under water stressed conditions. The normalization partially removed the disturbance caused by external factors such as soil interference, position of sun, illumination, and angle of view. That was not demonstrated in our study. Nonetheless, NWI-2 showed a marginal superiority over the two indices.

Association of SRI with Agronomic Traits

The association between the SRI and yield components was evaluated with the objective of determining if any particular yield component was driving the association with yield. The specific SRI that were best associated with yield (i.e., the NIR-based indices) also showed a greater correlation with these yield components. This observation is consistent with the fact that in all experiments grain yield, grains per square meter, and biomass showed significant positive correlations among themselves (data not presented). Waddington et al. (1986) and Sayre et al. (1997) clearly demonstrated that grain yield is particularly well correlated with grains per square meter in irrigated spring wheat in studies executed in the same location as used in this study. Therefore, it could be hypothesized that the relationship between NIR-based indices and grains per square meter may be the most important basis of the high relationship between NIR-based indices and grain yield.

Future Research Goals

Using an indirect selection tool is appropriate if the genetic correlation between the selected and unselected traits is very high, the heritability is much higher for the selected trait than for the unselected trait, and the correlated response in the unselected trait based on the selected trait is higher than the direct response to selection of the unselected trait (Falconer, 1989). In practice, this combination is rarely obtained. It is also important to consider the time and cost involved in using indirect selection tools compared to the use of grain yield per se as a selection criterion. A research project is currently underway to estimate heritability, expected genetic gain, correlated response to selection for grain yield estimated from the SRI, and the efficiency of selecting superior genotypes for grain yield based on SRI compared to the selection of superior genotype based on yield per se at different geographic locations and at different moisture conditions under the supervision of Oklahoma State University and CIMMTY, Mexico. Efforts are ongoing to develop a new lightweight spectral sensor to take measurements for these NIR-based indices in the field. The new sensor (approximate cost US$ 4000–5000) will facilitate faster measurements in the field compared to the spectroradiometer (cost US$ 30 000) used in the current studies and will bring down the current cost of equipment to a minimum level. These improvements should facilitate the adoption of this selection criterion by breeders and enhance their ability to discriminate genotypes for grain yield in the breeding trials.

CONCLUSIONS

Spectral reflectance indices have shown the potential to differentiate genotypes for grain yield in this study with different types of breeding lines of spring wheat under irrigated conditions. The best growth stages to apply the indices to differentiate genotypes for grain yield were heading and grainfilling. Comparing various indices, the indices based on NIR (WI, NWI-1, and NWI-2) demonstrated consistently higher levels of association and explained a higher proportion of the variability for grain yield compared with the other spectral indices (RNDVI, GNDVI, SR, and PRI). The NIR-based index (NWI-2) showed a very high efficiency in selecting superior genotypes in different experiments. The correlations in random populations confirm a genetic basis or link between yield and the physiological characteristics indicated by SRI.

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