https://doi.org/10.1590/1678-4499.20210067

# Correlation among vegetative and reproductive variables in wheat under a climate change simulation

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**Received:** Mar. 4, 2021 | **Accepted:** Jun. 7, 2021

**Section Editor:** Gabriel Constantino Blain

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**How to cite:** Argentel-Martínez, L., Peñuelas-Rubio, O., Ponce, J. A. L., Arredondo, T., Garatuza-Payan, J. and Yepez, E. A. (2021). Correlation among vegetative and reproductive variables in wheat under a climate change simulation. Bragantia, 80, e4221. https://doi.org/10.1590/1678-4499.20210067

**ABSTRACT:** Based on climate change scenarios predicted for northwestern Mexico, an experiment was carried out during the 2016–2017 and 2017–2018 crop cycles, under field conditions in wheat, in the Yaqui Valley, Sonora, Mexico. The assay consisted on canopy temperature increase by + 2 °C with respect to ambient temperature, using a temperature free-air controlled enhancement (T-FACE) system for temperature manipulation and control. This experiment aimed to determine the existing correlation among vegetative and reproductive variables that can result precise indicators of warming tolerance. A total of 30 variables divided into: morphological (6), physiological (7), biochemical (8) and agronomic (9) indicators were evaluated, using CIRNO C2008 cultivar as experimental model. For each variable, the response index, in a total of five repetition each year, was computed. Results indicated that, during the vegetative stages, the most precise variables for heat tolerance evaluation were: dry matter, vegetal vigor, water potential, nitrate reductase and transpiration indexes; while during the reproductive stage were: grain and biomass yield, spike mass and vain grain per spike indexes. There was a positive correlation among the most majority of vegetative and reproductive variables, being water and osmotic potential indexes those with the greatest contribution to biomass and grain yield. From the 30 evaluated variables, water and osmotic potential, transpiration, grain yield and field water use efficiency indexes were the most precise indicator of heat tolerance in CIRNO C2008 under canopy temperature increase in + 2 °C.

**Key words:** phenology, physiology, warming.

## INTRODUCTION

Heat stress has, nowadays, become one of the most important abiotic factors affecting agriculture worldwide (Asseng et al. 2015). Around 59% of the world agricultural area is affected by this type of stress, where, in most cases, heat intensity exceeds the tolerance indexes of traditional crop species (Kurepin et al. 2015).

In Mexico, the arid and semi-arid ecosystems cover 51% of the territory (Díaz-Padilla et al. 2011), where drought and salinity constitute the most common stresses. Additionally, the expected increases in global temperature impact these ecosystems (IPCC 2014). This factors combination exacerbates plants physiological and yield performance of several basic crops, such as wheat (Argentel-Martínez et al. 2019).

Warming is a common stress generally overlooked by producers (Rezaei et al. 2018); however, it modifies the initial development, vegetation index, water regime, transpiration rate, hormonal metabolism, photosynthesis-respiration balance, water use efficiency, protein synthesis, enzymatic activity (Thomason et al. 2018), time of phenophase occurrence (Garatuza-Payan et al. 2018) and, consequently, decreases grain and industrial yields (Yang et al. 2017). Asseng et al. (2015) reported that wheat will be one of the most affected crops by heat stress in the next decade. However, current wheat crop production worldwide still demonstrates its capacity to grow and produce under different climates (Argentel-Martínez et al. 2018). In the Yaqui Valley, Sonora, Mexico, more than 39% produce around 39% of the national wheat production every year. In this valley, a recent study of the initial response of wheat to experimental warming showed that phenology and grain yield decreased up to 30% due to heat stress (Garatuza-Payan et al. 2018).

The impact of heat stress in crops are generally, based on modeling and simulation, considering as a base the yield response variation for different years (through analysis of temperature variations). This approach is used because of the lengthy and time consuming that result the evaluation of indicators such as physiological and biochemical variables in field tests. Then, the evaluation of ecophysiological and agronomic variables, tested under field conditions, would contribute to understanding heat tolerance. Abiotic stress response should be explained under field conditions, where the broadest interaction genotype-environment can be observed. Although various studies of warming response of wheat have already been developed using some physiological or agronomic variables (Garatuza-Payan et al. 2018; Argentel-Martínez et al. 2019), the general response has not been fully described through variables integration (Rezaei et al. 2018).

During plants exposition to heat stress, several morphological, physiological, biochemical and agronomical mechanisms are activated, but not all contribute to general or final heat tolerance (Argentel-Martínez et al. 2018). For this reason, in order to provide more information about the tolerance degree of certain species to stressing conditions, it is important to analyze a large number of variables in both the vegetative and the reproductive stages and its correlation (Dettori et al. 2017). Therefore, this work aimed to evaluate the existing correlation among vegetative and reproductive variables in wheat in a climate change scenario based on temperature increase, under field conditions, in the crystalline wheat cultivar CIRNO C2008, and recommend variables as precise indicators of heat stress tolerance.

## METHODS

The experiment was carried out during the two growing seasons of 2016–2017 and 2017–2018, under field conditions at the Experimental Technology Transfer Center (CETT-910) of the Instituto Tecnológico de Sonora (ITSON), located in the Yaqui Valley at 27°22’0.4” N and 109°54’50.6” W (UTM: 607393.24 m east; 3027508.34 m north) at 46 m above sea level.

### Treatments and temperature control

The experiment consisted in raising crop canopy temperature by 2 °C, using the temperature free-air controlled enhancement (T-FACE) system proposed by Kimball (2015) and fully described by Garatuza-Payan et al. (2018). Two treatments were established to study the wheat response to increased temperature: increased canopy temperature aimed at 2 °C (T1: warming treatment) above ambient canopy temperature of adjacent plots (T2: control treatment). In both crop cycles, six equilateral triangular structures of 5.2 m of side where used (Fig. 1). Each structure was equipped with six thermal radiators per plot (FTE-1000 model, 1000 W, 240 V, 245 mm long × 60 mm wide, built by Mor Electric Company Heating Association Inc. Comstock Park, MI, USA). Two radiators were mounted in each triangle side, such that spatial distribution of radiators formed a regular hexagon that rose canopy temperature by 2 °C on 3 m diameter plots.

Temperature control was achieved with infrared temperature sensors (IRTS; Apogee Instruments, Inc., Logan, UT, USA) installed on both control and warmed plots with an inclination of 45° from the soil surface to cover an ellipsoid of r = 1.5 m at the center of the plot. The IRTS signal was received in a datalogger (CR1000 Campbell Sci, Inc., Logan, UT, USA). The electronic system was programmed to keep a constant temperature of 2 °C in the warming treatments, through the proportional, integrative and derivative routine described in Kimball (2015), simulating the temperature increase predicted for this region for the year 2050, according to the IPCC (2014).

**Figure 1.** Aerial image of the experimental warming assay during the crop cycles 2016–2017 and 2017–2018.

## Climate variables behavior

In both experimental seasons, average temperatures remained between 8 and 24 °C. The lowest temperatures occurred during December and January. Monthly rainfall was less than 25 mm in both years and the largest precipitation record occurred in February for the 2016–2017 campaign, and in March for 2017–2018. The wind speed did not exceed 3 m·s$^{-1}$ in both years (Fig. 2), an aspect that caused considerable stability of the T-FACE system and the thermal control of the crop canopy.

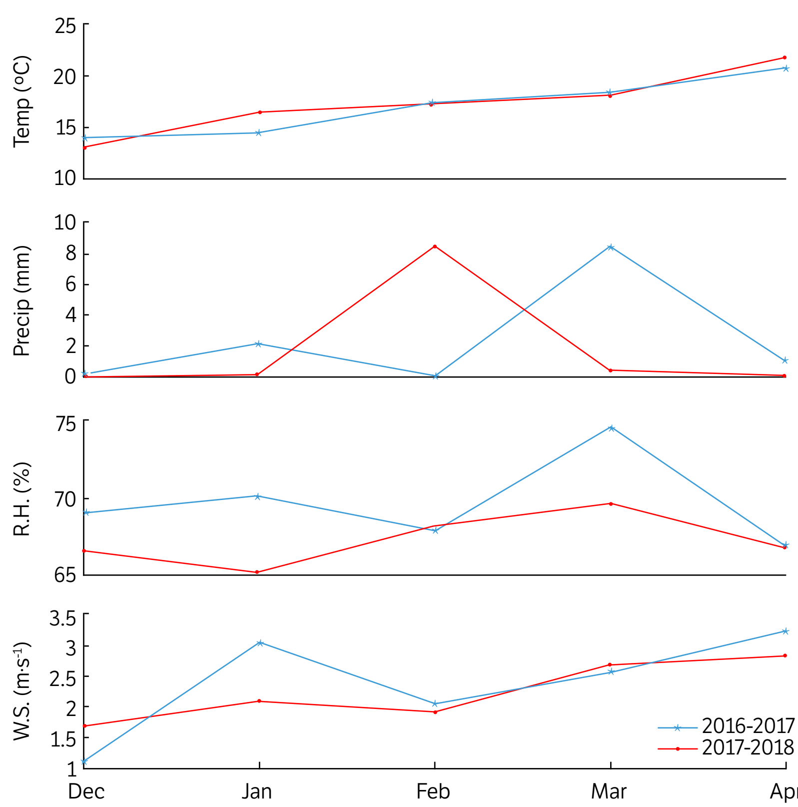


**Figure 2.** Climate variables during 2016–2017 and 2017–2018 crop cycles.

Note. WS: wind speed; R.H: relative humidity; Precip: precipitation; Temp: temperature.

## Cultivar used as a model, seeding and culture attentions

CIRNO C2008 wheat cultivar was used in both crop cycles due to its genetic stability since 2008 (Argentel-Martínez et al. 2018), when was commercially released in southern Sonora (Figueroa-López et al. 2010). This cultivar is classified as crystalline or hard wheat (*Triticum durum* L.). It was originated from selection in segregating populations of SOOTY-9/RASCON-37//CAMAYO crossbreed, carried out at the International Center for Maize and Wheat Improvement (CIMMYT). The average height is about 78 cm with erect stems. Under irrigation, physiological maturity is reached at 122 days. Mean grain yield reaches 5.6 and 6.3 $t \cdot ha^{-1}$. Every year and average of 150 000 ha of this cultivar are sowed in the Yaqui Valley (Garatuza-Payan et al. 2018).

Seeding was carried after tillage (Table 1) in a vertisol soil (Bockheim et al. 2014), with a sowing machine (SUB-24) with three rows on the furrows using a seeding density of 170 $kg \cdot ha^{-1}$. The background fertilization includes standard doses of 250 $kg \cdot ha^{-1}$ of urea + 100 $kg \cdot ha^{-1}$ of monoammonium phosphate fertilizer (MAP), 11–52–00 in both crop cycles. During the first and second irrigations, which took place during growth and tillering phenophases, respectively, nitrogen fertilizer was applied at a dose of 50 $kg \cdot ha^{-1}$ of urea. In the third irrigation (heading phenophase), there was no fertilizer application.

**Table 1.** Soil tillage during the two years of experimentation.

| Activities | Depth (cm) | Date | |
|---|---|---|---|
| | | 2016 | 2017 |
| Subsolation | 45 – 50 | 14/10 | 6/10 |
| Plowing | 20 – 25 | 18/10 | 10/10 |
| Double tillage | 10 – 15 | 21/10 | 14/10 |
| Terrain leveling | - | 25/10 | 17/10 |
| Furrowing | 20 | 28/10 | 20/10 |

All irrigations were applied with an average water depth of 14 cm and irrigation intervals of about 25 days, when gravimetric water volume of the soil was at about 70%.

During the growing period the presence of foliage aphid (*Schizaphis graminum*) was detected, which was controlled with Muralla Max pesticide (a.i. imidacloprid + beta-cyfluthrin) at a dose of 0.20 $L \cdot ha^{-1}$ on the periphery of the plot to 2–3 m from the border of the experimental area. A slight presence of broadleaf weeds was observed and they were controlled by manual weeding before applying irrigations.

## Evaluated variables

The evaluated variables were determined using the respective methodologies and are summarized in Table 2.

All variables were expressed as indexes related to the control, using the following equation (Eq. 1):

$$IV(\%) = \frac{VW}{VC} \text{ x } 100 \quad (1)$$

where VI represents the index of the variables, expressed in %; and VW and VC represent evaluated variables under warming and control plots, respectively.

The variables nomenclature was formed by the using of the word "index" after variable name: initial plant height index (PHI); root length index (RLI); fresh matter accumulation index (FMI); dry matter accumulation index (DMI); vegetal vigor index (VVI); water potential index (WPI); osmotic potential index (OPI); osmotic adjustment index (OAI); proline content index (PI); glycine betaine content index (GBI); reduced glutathione activity index (RGI); POD activity index (PODI); nitrate reductase activity index (NRI); glutamine synthetase activity index (GSI); protein content during flowering index (TPCI); starch content during flowering index (TSCDFI); NDVI index (NDVII); photosynthesis index (PHOTOI); transpiration index (TRANI); leave water use efficiency index (LWUEI); phenology index (PHENI); spike length index (SPIKELI); spike mass index (SMASSI); full grain per spike index (FG/SI); vain grain per spike index (VG/SI); grain mass index (GMI); biomass yield index (BIOYI); grain yield (GYI); starch mobilization rate index (SMI) and field water use efficiency index (FWUEI).

**Table 2.** Response variables, units, acronyms, type of variable and method used for measurements.

| Variables | Units | Acronyms | Type | Methodology |
|---|---|---|---|---|
| **Vegetative stages** | | | | |
| Initial plant height | cm | PHI | Mor | Foroughbakhch-Pournavab et al. (2015) |
| Root length | cm | RLI | Mor | |
| Fresh matter accumulation | g | FMI | Mor | |
| Dry matter accumulation | $mg{\cdot}g{\cdot}DM^{-1}$ | DMI | Mor | |
| Vegetal Vigor | adimensional | VVI | Morp | |
| Water potential | MPa | WPI | Phy | Blum et al. (2017) |
| Osmotic potential | MPa | OPI | Phy | |
| Osmotic adjustment | MPa | OAI | Phy | Blum et al. (2017) |
| Proline content | $\mu g{\cdot}g{\cdot}FM^{-1}$ | PI | Bio | Bates et al. (1973) |
| Glycine betaine content | $\mu g{\cdot}g{\cdot}FM^{-1}$ | GBI | Bio | Grieve and Grattan (1983) |
| Reduced glutathione activity | $\mu mol{\cdot}g{\cdot}FM^{-1}$ | RGI | Bio | Xue et al. (2001) |
| POD activity | $EAU{\cdot}g{\cdot}FW^{-1}$ $min^{-1}$ | PODI | Bio | Martínez (2001) |
| Nitrate reductase activity | $\mu mol\ NO_2^-{\cdot}g{\cdot}FW^{-1}{\cdot}min^{-1}{\cdot}h^{-1}$ | NRI | Bio | Jaworsky (1971) |
| Glutamine synthetase activity | $\mu mol{\cdot}GH{\cdot}s^{-1}{\cdot}g^{-1}$ FW | GSI | Bio | Slawky and Rodier (1988) |
| Protein content during flowering | $mg{\cdot}g{\cdot}DM^{-1}$ | TPCI | Bio | Yoshida et al. (1971) |
| Starch content during flowering | $mg{\cdot}g{\cdot}DM^{-1}$ | TSCDFI | Bio | McCready et al. (1950) |
| NDVI | adimensional | NDVII | Phy | Inman et al. (2005) |
| Photosynthesis | $\mu mol\ CO_2{\cdot}m^{-2}{\cdot}s^{-1}$ | PHOTOI | Phy | Yang et al. (2016) |
| Transpiration | $\mu mol\ H_2O{\cdot}m^{-2}{\cdot}s^{-1}$ | TRANI | Phy | |
| Leave water use efficiency | $\mu mol\ CO_2{\cdot}H_2O{\cdot}m^{-2}{\cdot}s^{-1}$ | LWUEI | Phy | Argentel-Martínez et al. (2018) |
| Phenology | days | PHENI | Mor | Zadocks et al. (1974) |
| **Reproductive stage** | | | | |
| Spike length | cm | SPIKELI | Agro | Garatuza-Payan et al. (2018) |
| Spike mass | g | SMASSI | Agro | |
| Full grain per spike | unit | FG/SI | Agro | |
| Vain grain per spike | unit | VG/SI | Agro | |
| Grain mass | g | GMI | Agro | |
| Biomass yield | $t{\cdot}ha^{-1}$ | BIOYI | Agro | |
| Grain Yield | $t{\cdot}ha^{-1}$ | GYI | Agro | |
| Starch mobilization rate | % | SMI | Bio | Yang et al. (2002) |
| Field water use efficiency | $m^3\ H_2O{\cdot}t^{-1}{\cdot}ha^{-1}$ | FWUE | Agro | Jin et al. (2018) |

Note. Mor = morphological; phy = physiological; bio = biochemical; agro = agronomical.

## Statistical processing

Data corresponding to response variables were used to form a 5 × 30 matrix, which were previously tested for normality and homoscedasticity (Svantesson and Wallace 2003). In order to determine variables with the greatest contribution to the total variability response in each stage, as criteria for assessing heat tolerance, a principal components analysis (PCA) was carried out (Pearson 1901). In this analysis, variables were selected when the value of its unitary contribution was higher than 0.8 (80%) (Shabala and Munns 2017).

Based on variables with the greatest contribution to the total variability (unitary contribution > 0.85), an analysis of hierarchical conglomerates of complete linkage was done, based on a Euclidean distance matrix (Rohlf and Fisher 1968), to group variables. The average tolerance values of the computed indexes of each group and the distance values between groups were determined. The correlation matrix obtained through a canonical correlation analysis was accomplished (Mertler and Reinhart 2016). For the statistical processing, STATISTICA professional statistical package, version 8.4, was used (StatSoft, Tulsa, OK, USA).

## RESULTS AND DISCUSSION

Through the PCA, developed to the vegetative variables, it was found that among all evaluated variables, 69.93% of the total variability was accumulated in the first two components in the vegetative stages. The variables FMI, DMI, VVI, WPI, SPI, GBI, WPI, OPI, NRI, HSCI, NDVII and TRANI were the ones with greater contribution (46.21%) in the first component, while PHI, PHOTOI and LWUEI contributed with 23.72% to the second component (Table 3).

**Table 3.** Principal components analysis of the variables during the vegetative stages (marked loadings are > 0.7).

| Variables | Components | |
|---|---|---|
| | 1 | 2 |
| PHI | –0.147532 | –0.798777 |
| RLI | –0.447731 | 0.631476 |
| FMI | 0.917772 | 0.357437 |
| DMI | 0.929419 | 0.280641 |
| VVI | 0.953872 | 0.168694 |
| WPI | 0.869861 | –0.054318 |
| OPI | 0.960113 | 0.049827 |
| OAI | 0.155628 | 0.448907 |
| PI | –0.006277 | 0.549976 |
| GBI | 0.858650 | 0.342389 |
| RGI | 0.860899 | –0.329082 |
| PODI | 0.615439 | –0.645297 |
| NRI | 0.756755 | –0.162033 |
| GSI | 0.673860 | –0.291595 |
| TPCI | –0.152677 | 0.066192 |
| TSCDFI | 0.757495 | –0.513911 |
| NDVII | –0.764545 | –0.52598 |
| PHOTOI | 0.193311 | –0.920749 |
| TRANI | 0.865616 | –0.322563 |
| LWUEI | –0.409009 | –0.902483 |
| PHENI | –0.378921 | –0.366832 |
| Expl. var | 9.704084 | 4.98043 |
| Contribution | 0.462099 | 0.237163 |
| Total contribution percentage | 69.93% | |

The present study confirms the contribution of the initial development (FMI, DMI, VVI), the water regime (WPI, OPI) and the accumulation of osmotically active compounds (GBI and RGI) variables to the physiological performance of the plants. Such indicators have been reported as efficient variables for the heat response in wheat (Bharati et al. 2018; Dwivedi et al. 2018). These variables of water and osmotic regime allowed a significant contribution of transpiration (TRANI) and water use efficiency (LWUEI) to the total variability found during the vegetative stage. Special attention has been given to the accumulation of osmotically active compounds, mainly glycinebetaine and reduced glutathione, which represent a biochemical signal of abiotic stress in plants (El Sabagh et al. 2018).

On the other hand, the activity of some enzymes, such as nitrate reductase (NRI), contributes to the variability of the responses of the species in the initial stages and development, as well as in the present study. This result has shown that under heat conditions, the efficiency of nitrogen fertilizer assimilation is increased (Bala and Sikder 2018). Also, high nitrogen assimilation contributes to a grader photosynthetic activity due to the chlorophyll increase still under heat stress (Kocheva et al. 2020).

For the reproductive stage, SMASSI, BIOYI, SMI, GYI and FWUE showed the most contribution to the first component, contributing with 53.41% of the total variability. In the second component VG/SI was the only variable with a significant contribution, which contributed in a 23.8% to the total variability (Table 4).

**Table 4.** Principal components analysis of the variables during the reproductive stages (Marked loadings are > 0.70).

| Variables | Components | |
|---|---|---|
| | 1 | 2 |
| SPIKELI | 0.641452 | −0.418583 |
| SMASSI | 0.899695 | −0.194620 |
| FG/SI | −0.239384 | 0.261225 |
| VG/SI | −0.178851 | 0.848298 |
| GMI | 0.484230 | 0.784996 |
| BIOYI | −0.925951 | −0.184165 |
| GYI | 0.990337 | −0.068262 |
| SMI | −0.725711 | −0.630377 |
| FWUE | 0.947117 | −0.298447 |
| Expl. var | 4.806522 | 2.142178 |
| Contribution | 0.534058 | 0.238020 |
| Total contribution percentage | 77.20% | |

## Correlation among variables during vegetative and reproductive stages

When the correlation among reproductive and vegetative stages variables were determined (Table 5), it was obtained that: SMASSI correlated negatively and significantly with FMI, DMI, VVI, WPI, and GBI. The variable VG/SI correlated negatively with GSI. The GMI variable correlated positively with RLI and negatively with TSCDFI and TRANI. The variable BIOYI correlated positively with VVI, OPI and negatively TRANI. The GYI correlated negatively with VVI, OPI, RGI and TRANI. The SMI correlated positively with WPI and with TSCDFI, and finally, FWUEI correlated positively with OPI.

The spikes mass (SMASSI) is one of the variables that most contribute to grain yield and is determined by the amount of filled grains (FG/SI). Both components are function of plant capacity to mobilize elaborated substances from the source (leaves and stem) to the sink (the grain) (Ram et al. 2018), which decreases as a result of heat effect because of water regime variability. In the present study, the spike mass of the (SMASSI) was affected by the variable water regime (WPI and OPI) and by the accumulation of osmotically active compound GBI. The synthesis of osmotically active compounds involves the use of carbon skeletons or the degradation of macromolecules for their synthesis. Perhaps these organic compounds were degraded to form glycine betaine, whose main role in plants under stress condition is to increase osmotic pressure to reduce water potential and ensure transpiration (Argentel-Martínez et al. 2019), perhaps for mesophyll cooling. Some variables of the vegetative stage contribute to grain quality and that the responses in yield components could be attributed to variations during the vegetative stage, where a high cellular dynamism take place (Yáñez et al. 2017).

**Table 5.** Correlations coefficient of variables during vegetative stage (first column) and reproductive stage (first row).

| Variables | SPIKELI | SMASSI | FG/SI | VG/SI | GMI | BIOYI | GYI | SMI | FWUE |
|---|---|---|---|---|---|---|---|---|---|
| PHI | –0.19 | 0.48 | –0.79 | –0.37 | –0.52 | 0.12 | 0.09 | 0.14 | 0.34 |
| RLI | –0.20 | 0.13 | 0.27 | 0.60 | 0.92* | –0.51 | 0.26 | –0.81 | 0.04 |
| FMI | –0.10 | –0.83* | 0.58 | –0.27 | –0.09 | 0.63 | –0.70 | 0.49 | –0.70 |
| DMI | –0.22 | –0.83* | 0.50 | –0.24 | –0.12 | 0.69 | –0.76 | 0.49 | –0.74 |
| VVI | –0.21 | –0.88* | 0.30 | –0.17 | –0.25 | 0.84* | –0.83* | 0.56 | –0.78 |
| WPI | –0.32 | –0.83* | 0.62 | –0.10 | –0.47 | 0.67 | –0.79 | 0.83* | –0.79 |
| OPI | –0.42 | –0.95** | 0.51 | –0.01 | –0.35 | 0.83* | –0.92** | 0.71 | –0.92* |
| OAI | 0.23 | –0.34 | –0.15 | 0.24 | 0.26 | 0.28 | –0.15 | –0.21 | –0.20 |
| PI | 0.53 | 0.21 | 0.47 | –0.59 | 0.43 | –0.46 | 0.40 | –0.25 | 0.35 |
| GBI | –0.12 | –0.94** | 0.45 | 0.05 | –0.12 | 0.74 | –0.77 | 0.49 | –0.81 |
| RGI | –0.52 | –0.77 | –0.07 | –0.02 | –0.59 | 1.00** | –0.93** | 0.71 | –0.80 |
| PODI | –0.72 | –0.46 | –0.01 | –0.05 | –0.70 | 0.71 | –0.75 | 0.75 | –0.61 |
| NRI | –0.21 | –0.44 | 0.36 | –0.57 | –0.42 | 0.50 | –0.54 | 0.65 | –0.42 |
| GSI | 0.17 | –0.24 | 0.05 | –0.85* | –0.62 | 0.48 | –0.33 | 0.70 | –0.13 |
| TPCI | –0.78 | –0.08 | –0.13 | 0.68 | 0.44 | 0.06 | –0.24 | –0.45 | –0.31 |
| TSCDFI | –0.18 | –0.54 | 0.18 | –0.38 | –0.87* | 0.70 | –0.64 | 1.00** | –0.50 |
| NDVII | 0.16 | 0.78 | –0.81 | 0.09 | –0.15 | –0.40 | 0.59 | –0.33 | 0.69 |
| PHOTOI | –0.51 | –0.06 | –0.64 | 0.04 | –0.82* | 0.59 | –0.43 | 0.55 | –0.22 |
| TRANI | –0.52 | –0.78 | –0.06 | –0.02 | –0.59 | 1.00** | –0.93** | 0.71 | –0.80 |
| LWUEI | –0.24 | 0.51 | –0.73 | 0.05 | –0.58 | –0.03 | 0.17 | 0.16 | 0.33 |

Marked correlations coefficient are significant (*) at $p < 0.05$ and highly significant (**) for $p < 0.05$.

The same variables (VVI, OPI, GRI and TRANI) which correlated positively with the BIOYI, affected in the GYI, but negatively. For BIOYI, the plant vigor (VVI) from the imposition of heat had a positive effect, and the reductions of the osmotic potential (OPI) and the GRI content, as a promoter of the osmotic potential variation, contributed to maintain the sufficient transpiration that allow the normal development of the plants (BIOYI); however, these contributions were not enough to contribute to the grain yield (GYI).

Heat stress cause an increase in the quantity of vain grains, mainly those of the distal part of the spike (Chandra et al. 2017) causing a reduction of grain mass (SMASSI), as occurred in the present study.

## Clusters of indicators of heat variability response

Three groups were conformed from variables with the greatest contribution to the total variability (Fig. 3). Group 1 included FWUE, GYI, BIOYI and SMASSI, being the most efficient variables to evaluate warming response variability, based on its highest indices (Table 6), all of them belonging to agronomic indicators.

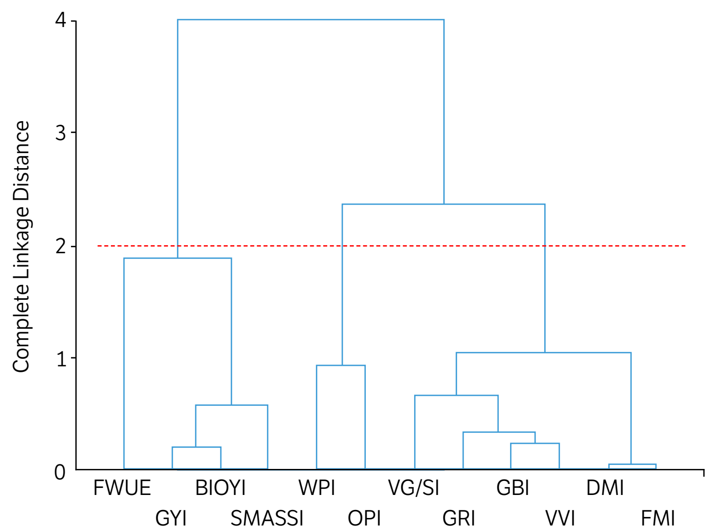


**Figure 3.** Cluster of indices of the variables of greatest contribution and correlation among vegetative and reproductive stages.

**Table 6.** Mean values of indexes of variables by group and distance values between groups.

| Group | Indexes | Distances from respective cluster center | Distance among groups | |
|---|---|---|---|---|
| | | | 1 | 2 |
| I | FWUE | 0.57 | | |
| | GYI | 0.05 | | |
| | BIOYI | 0.09 | | |
| | SMASSI | 0.22 | | |
| II | WPI, | 0.18 | 0.67 | |
| | OPI | 0.19 | | |
| III | VG/SI | 0.16 | 1.35 | 0.69 |
| | GBI | 0.02 | | |
| | GRI | 0.02 | | |
| | VVI | 0.12 | | |
| | DMI | 0.16 | | |
| | FMI | 0.20 | | |

The second group was formed by only two variables: WPI and OPI, both are physiological indicators evaluated during the vegetative stage. The third group clustered VG/SI (agronomical indicator from reproductive stage), GRI, GBI (biochemical indicators) and VVI, DMI, and FMI (morphological indicators), all of the variables evaluated during vegetative stage, which resulted in less accurate indicators to evaluate responses to heat stress in crystalline wheat. Ashraf and Foolad (2007) pointed out that among the osmoregulators synthetized by plants, glycine betaine (GB) and reduced glutathione (RGI) are a precise biochemical indicator in improving plant abiotic stress resistance.

When average values of Euclidian distance from the cluster center were calculated for the variables agglutinated in each group, it was found that yield components indexes (group 1) were those of lower values, demonstrating the existence of positive correlation among them. The GYI showed the minimum value of distance (Table 6).

The second group, formed by water regime variables, had an intermediate average value, while the third group presented the highest values, being the least accurate for the identification of heat response variability. Groups 1 and 2 are the most effective groups of variables for heat variability response of the evaluated cultivar due to their lower Euclidian distance.

Ever since decades ago, scientists have worked on the use of indicators as criteria for variability to heat response during genotype valuation (Elbashir et al. 2017). Many of these indicators are common to different types of stress, such as salinity, drought and heat. For heat stress in particular, grain yield, thousand grains weight, grain filling duration, number of effective full and vain grain per plant (Hunt et al. 2018) and biomass (Hütsch et al. 2018) are some of the most important yield components indicators of variability response to heat. In this work, GYI, BIOI and SMASSI were the variables with major contribution to agronomic variability. However, according to natural intra e interspecific plant response variability to abiotic stress, it is very important to monitor the available germplasm through morphological, physiological, biochemical and yield components indicators in order to elucidate the most precise indicators in some crops, including wheat (Thomason et al. 2018). In some cases, due to the costs of agronomic experimentation for abiotic stress under field conditions, an important alternative is the use of elite varieties that have maintained their productive genetic potential over the years, such as CIRNO C2008 (Argentel-Martínez et al. 2019). That is why it was used as experimental model in this work.

Generally, spike mass has been correlated with its size, however other researchers have rejected this correlation since the spike can grow according to its genetic potential but not to fill all of its grains, reducing in consequence its mass, and backwards (Garatuza-Payan et al. 2018).

Plant water relation variables have always been associated to stress due to drought and salinity (Argentel-Martínez et al. 2018). However, in this study, WPI and OPI were efficient indicators of heat response variability with respect to the control. Other physiological indicators have also been proposed for heat stress tolerance, such as photosynthetic rate index. However, in this study, a high average photosynthetic activity (PHOTOI) was not an accurate indicator, because it was associated with high transpiration (TRANI), creating significant variability in the efficiency of leaf water use (LWUEI) and field water use efficiency (FWUEI) (Hütsch et al. 2018).

The TSCI has shown association with yield in warm environments, denoting its association with heat stress tolerance (Allahverdiyev et al. 2018). The TSCI shows high correlation with yield and have been an efficient indicator for breeding for warming tolerance in wheat. Anyway, genotypes having cooler canopies shows greater grain filling and consequently maintain lower reduction of SMASSI (Fan et al. 2018).

## CONCLUSION

The variables of the initial growth (FMI, DMI, VVI), water regime (WPI, OPI) and osmotically active accumulation (GBI and RGI) were the ones of major contribution to the total variability of reproductive stage variables. Vegetative variables contributed in a 69% to the total variability found.

The variables SMASSI, GMI, BIOYI and GYI of the reproductive stage were the ones of major correlation with vegetative stages variables. Reproductive stage variable explained 77% of total variability.

The most precise variables for the warming response variability in the studied cultivar were: SMASSI, BIOYI and GYI, followed by water regime variables WPI, OPI and the osmotically active accumulation variables GRI and GBI.

## AUTHORS' CONTRIBUTION

**Conceptualization:** Argentel-Martínez L.; **Methodology:** Yepez E. A. and Argentel-Martínez L.; **Investigation:** Garatuza-Payan J. and Peñuelas-Rubio O.; **Writing – Original Draft:** Argentel-Martínez L.; **Writing – Review and Editing:** Yepez E. A. and Leyva P. A.; **Funding Acquisition:** Arredondo T., Garatuza-Payan J and Argentel-Martínez L.; **Resources:** Arredondo T., Garatuza-Payan J and Argentel-Martínez L.; **Supervision:** Arredondo T., Garatuza-Payan J and Argentel-Martínez L.

## DATA AVAILABILITY STATEMENT

Data sharing is not applicable.

## FUNDING

Consejo Nacional de Ciencia y Tecnologia
https://doi.org/10.13039/501100003141
Grant No. 220788

Consejo Estatal de Ciencia y Tecnología - Sonora
Grant No. 20190430

## ACKNOWLEDGMENTS

To TecNM and ITSON.

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