Environmental and Experimental Botany 224 (2024) 105825

Contents lists available at ScienceDirect

Environmental and Experimental Botany

journal homepage: www.elsevier.com/locate/envexpbot

Interspecies diversity in morphological responses to water stress: Study on a panel of weed and crop species

Quentin Cournault[a,b,c], Nathalie Colbach[a], Hugues Busset[a], Annick Matejicek[a], Pauline Souche-Suchovsky[a], Marion Prudent[a], Delphine Moreau[a,*]

[a] *Agroécologie, INRAE, Institut Agro, Univ. Bourgogne, Univ Bourgogne Franche-Comté, Dijon 21000, France*
[b] *Biogéosciences, UMR 6282 CNRS, Université de Bourgogne, 6 boulevard Gabriel, Dijon 21000, France*
[c] *AgroParisTech, Palaiseau 91123, France*

ARTICLE INFO

Keywords:
Biomass allocation
Climate change
Competition
Plasticity
Specific leaf area
Trait

ABSTRACT

Climate change modifies the dynamics and the quantity of plant water supply, and plant morphological response to environmental factors plays a key role in crop-weed interactions. This study investigated the interspecies diversity related to the morphological responses of annual herbaceous species to water stress. Key morphological traits were measured at two growth stages on five weed and two crop (soft wheat, rapeseed) species grown on a gradient of water availability in a greenhouse experiment. For each trait, response curves to water stress were defined, and their parameters were used to quantify interspecies diversity. Generic morphological response patterns were identified across all species and plant stages. Water stress reduced leaf area per unit leaf biomass (SLA, lowering water demand) and increased the ratio of plant height to aboveground biomass for all species (HBR, keeping access to light). In most situations, the ratio of root biomass over total biomass (RBR) increased (improving water uptake). Variability in parameter values of morphological traits was primarily explained by the species, followed by growth stage. *Geranium dissectum* L. and *Abutilon theophrasti* Medik. were the most responsive species to water stress, especially at the flowering stage, with strong RBR and HBR increase and slight SLA decrease. Species differences were not related to clade (monocotyledonous/dicotyledonous) nor status (weed/crop), despite a near-significant clade effect on allocation of aboveground biomass, with grass species allocating more biomass to stems (vs leaves), while no general tendency was observed in broadleaved species. These findings provide new insights on comparative ecology of weed and crop species response to water limitation, and more research is expected to cover a wider range of weed and crop species.

1. Introduction

Among pests of arable crops, weeds are the main cause of yield losses, as they compete with the crops for resources (light, minerals and water) (Oerke, 2006). With climate change, competition for water will increase as (1) water is becoming ever scarcer in the soils because of changes in rainfall patterns and droughts, and (2) rising temperatures will increase plant transpiration and therefore plant demand for water. This situation will modify crop-weed competition for water in agroecosystems (Gleick, 1989; Ramesh et al., 2017).

Plant morphology is important for crop-weed interactions, as it determines (1) how a plant occupies the surrounding space and (2) which resource pools the plant can explore, sometimes to the detriment of neighbouring plants, of the same or a different species (Colbach et al., 2019). Under water stress, species present varying degrees of morphological plasticity (e.g., changing biomass allocation to leaves, stems or roots), and drought events thus reshape the spatio-temporal aboveground and belowground growth of plants in a field. This redefines the extent of the resource pools to which the plants have access for their growth.

Previous work on the morphological responses of crop and weed species to water stress focused (1) on only one or two species (Chahal et al., 2018; Monaco et al., 2005) and (2) on the germination and emergence phases (Singh et al., 2022). Current literature does not cover the diversity of species in arable cropping systems, especially for weeds and at different phenological stages. Such knowledge is though needed to predict future crop-weed interactions under limiting water conditions, and more broadly which species will survive and reproduce.

---

[*] Correspondence to: Delphine Moreau, INRAE, 17 rue Sully, Dijon 21000, France.
E-mail address: delphine.moreau@inrae.fr (D. Moreau).

https://doi.org/10.1016/j.envexpbot.2024.105825
Received 8 February 2024; Received in revised form 16 May 2024; Accepted 19 May 2024

Field trials are not adapted to accurately measure the single effect of water stress on crop and weed morphology, because of limited environmental control (difficulties to set levels of water limitation and to isolate the effect of water stress on morphology). Greenhouse experiments tried to overcome these shortcomings and studied plant morphological behaviour in response to water stress with regular manual watering/monitoring (Chahal et al., 2018; Monaco et al., 2005). However, this approach is time-consuming, with a high risk of handling errors (approximate volume of water added, insufficient monitoring frequency…). To avoid such issues, recent greenhouse studies worked with automatic watering platforms (Li et al., 2021; Ma et al., 2019; Moreau et al., 2022).

To our knowledge, the work of Moreau et al. (2022) is to date the only one that (1) studied the morphological responses of the largest number of weed species (three species) to soil-water availability (five levels) at vegetative and flowering stages, (2) with an automatic watering platform for precise monitoring, so that plants were placed on a conveyor belt and automatically weighed (and possibly watered) to keep close to the targeted soil-water availability throughout the experiment. With reduced water availability, all tested species were shown to produce smaller leaves per unit leaf biomass and taller plants per unit aboveground biomass. Some species increased (1) aboveground biomass allocation to leaves to the detriment of stems and/or (2) biomass allocation to roots to the detriment of aboveground plant compartments. This pioneer work though did not assess sufficient species for comparative ecology.

Consequently, our objectives were to investigate further species to (1) identify general responses patterns of morphological traits to water limitation, (2) investigate the interspecies diversity in plant morphological responses to water limitation at several growth stages, (3) explain this diversity in terms of species characteristics and (4) identify categories of morphological behaviours.

We thus conducted a greenhouse experiment with an automatic watering platform on seven annual herbaceous species (five weed and two crop species), all commonly found in arable fields and under temperate climate, and confronted with more water availability levels than in the work of Moreau et al. (2022) (seven levels). Species were compared on the basis of the parameters of the response curves of morphological variables to water stress. To our knowledge, this approach has never been used to assess morphological responses of weeds to water stress, although model parameters are relevant criteria for comparing intrinsic behavioural differences between genotypes (Tardieu, 2003) and species (Moreau et al., 2017).

Species response was assessed through four key morphological traits involved in crop-weed competition for light and thus in weed harmfulness for crop production (Colbach et al., 2019), already used in the work of Moreau et al. (2022).

Two traits describe how plants allocate biomass to different compartments (biomass-allocation traits):

- Root biomass to total plant biomass ratio (RBR) reflects biomass allocation to roots and therefore the propensity to explore soil to take up water and other soil resources to the detriment of light interception. The higher its value is, the more plants favour root over aboveground growth.
- Leaf biomass to aboveground biomass ratio (LBR) reflects the efficiency of leaf production (and therefore light interception) from aboveground biomass. Plants with more leaves (and therefore more light interception area) have larger LBR values.

Two other traits describe how biomass is transformed into size (size-related traits):

- Plant height to aboveground biomass ratio (HBR) reflects the ability to grow taller from a given aboveground biomass and, therefore, to dominate the canopy for light interception. The larger HBR is, the taller the plants are for a given aboveground biomass.
- Leaf area to leaf biomass ratio, also called specific leaf area (SLA) reflects the efficiency to produce leaf area (for light interception) from a given leaf biomass. High values indicate large leaves per unit leaf biomass whereas low values mean smaller leaves per unit leaf biomass.

2. Material and methods

2.1. Principle

A greenhouse experiment was carried out on an automatic watering platform of INRAE Dijon (France) in 2021 (4PMI, LemnaTec GmbH, Würlesen, Germany, https://plateforme4pmi.dijon.hub.inrae.fr/). Pre-germinated seeds of seven species (Section 2.2.1) were sown into irrigated pots to ensure emergence. Plants were then transferred on the conveyor belt for automatic watering, and each plant was assigned one out of seven water-availability levels (Section 2.2.2). Plants of each species were collected through destructive samplings, measured, and the four morphological traits presented in the introduction (Section 1) were calculated (Section 2.2.3).

For each plant, a water stress index was calculated (Section 2.3.1). Response curves of each morphological trait to the water stress index were fitted, and a species- and sampling stage-inherent parameter (assessing species × stage responses to water stress) was estimated (Section 2.3.2). Species parameter values were then compared to assess the effect of species and stage on morphological response to water stress (Section 2.3.3). To assess the weight and the drivers of the species effect on variability in plant morphology, morphological traits were analysed as a function of experimental factors, and then of species characteristics (Section 2.3.4). Finally, stress response parameters were used to build a species typology for morphological responses to water stress (Section 2.3.5).

2.2. Experiment

2.2.1. Species

Seven annual species were used in the experiment (Table 1): two crop species (soft winter wheat, *Triticum aestivum* L. – TRZAW and rapeseed, *Brassica napus* L. - BRSNN) and five weed species (*Abutilon theophrasti* Medik. - ABUTH, *Alopecurus myosuroides* Huds. - ALOMY, *Avena fatua* L. - AVEFA, *Geranium dissectum* L. - GERDI, *Tripleurospermum inodorum* (L.) CH Schultz - MATIN). The species were chosen according to several criteria: (1) be frequently found and co-exist in similar arable cropping systems and pedoclimates, (2) cover a diversity of families, belonging to the two clades (three monocotyledonous and four dicotyledonous species) and (3) have varied preferential emergence seasons (Table 1). All tested species grow on cool and moderately moist soils, on the basis of their Ellenberg moisture index. This index ranges from 1 (for species found in dry environments) to 12 (for plants usually submerged) (Hill et al., 1999).

2.2.2. Growth conditions

Seeds of all species were placed on Petri dishes in climate-controlled chambers, on moist filter paper for 5 days (temperatures of 20 °C to 30 °C depending on the species, 16 h photoperiod) to stimulate germination. On 12 March 2021, seedlings at the cotyledon stage were transplanted into 1 L individual pots (one plant per pot), made up of 540 g of inert clay substrate (80 % volume with expanded clay and 20 % with attapulgite, a fibrous clay). Pots were placed on fixed tables for 25 days, and automatically fertirrigated 5 times a day with a nutrient-rich solution (10% N – 10 % K – 10 % P), so that seedlings never underwent water stress.

At the 3–4 leaf stage (6 April 2021), 35 pots of ALOMY, 56 pots of ABUTH and 70 pots of all others species (Supplementary material A) were transferred to the automatic watering platform. Pots of each species were equally distributed across seven levels of water availability,

Table 1
Characteristics of the tested species.

EPPO code	Scientific name of the species	Clade	Family	Status	Season	Ellenberg moisture index (Hill et al., 1999)
ABUTH	*Abutilon theophrasti* Medik.	Dicotyledonous	Malvaceae	Weed	**Sp** - Su	4
ALOMY	*Alopecurus myosuroides* Huds.	Monocotyledonous	Poaceae	Weed	**A** – W - Sp	5
AVEFA	*Avena fatua* L.	Monocotyledonous	Poaceae	Weed	A – W - **Sp**	5
BRSNN	*Brassica napus* L. (rapeseed, cv Kador)	Dicotyledonous	Brassicaceae	Crop	**A**	4
GERDI	*Geranium dissectum* L.	Dicotyledonous	Geraniaceae	Weed	**A** - Sp	5
MATIN	*Tripleurospermum inodorum* (L.) CH Schultz	Dicotyledonous	Asteraceae	Weed	A – W – Sp - Su	5
TRZAW	*Triticum aestivum* L. (soft winter wheat, cv Calumet)	Monocotyledonous	Poaceae	Crop	**A** - W	5

The EPPO code (https://gd.eppo.int/) is a European identifier for the main crop pests. Species can emerge over one or several seasons (from Gardarin et al., 2010), abbreviated as follows: A = Autumn; W = Winter; Sp = Spring; Su = Summer. The preferred season of emergence is written in **bold**. All tested species co-exist in similar environments and grow on cool, moderately moist soils (according to their Ellenberg moisture indexes).

corresponding to 10 %, 20 %, 30 %, 40 %, 55 %, 75 % and 95 % (considered as optimal water supply) of field capacity. Water availability was defined as the transpirable fraction of soil water (FTSW), i.e. the ratio of available soil water to total transpirable soil water for a plant in a soil (Ramos and Martínez-Casasnovas, 2014)**.** Plant species and water treatments were randomly placed in the greenhouse.

The automated platform consists of conveyors with computer-controlled plant circulation (LemnaLauncher software bundle; LemnaTec, GmbH, Würlesen, Germany), that automatically drove plants three times a day to a watering unit (weighing terminal ST-Ex; Bizerba, Balingen, Germany and high-precision pump-watering station 520Du; Watson Marlow, Wilmington, MA, USA), without interruption until the end of the experiment (Jeudy et al., 2016; Moreau et al., 2022). In order to keep as close as possible to the targeted FTSW, pots were fertirrigated with a nutrient-rich solution (10 % N – 10 % K – 10 % P) only in case of insufficient water content, so that they reach the targeted FTSW again. As water supply never exceeded field capacity, water never drained out of the pots.

Throughout the experiment, artificial light was provided using 400 W lamps (HPS Plantastar; OSRAM, Munich, Germany), in addition to natural light. Mean photoperiod was 14.0 ± 1.3 h. Incident photosynthetically active radiation was 16.1 ± 2.5 mol$\bullet$m$^{-2}\bullet$day^{-1} (measured with silicon sensors; Solems, Palaiseau, France). Air temperature was 19.2 ± 1.1 °C (measured with PT100 sensors; Pyrocontrole, Meyzieu, France).

2.2.3. Plant measurements

For each species × water treatment combination, four plants of ABUTH and five plants of each other species were sampled at each of the following phenological stages: (1) 'early vegetative' for plants of all species and (2) excepted for ALOMY, either 'late vegetative' (AVEFA, BRSNN and TRZAW, which did not flower during the experiment) or 'flowering' (ABUTH, GERDI and MATIN).

The first sampling session was performed 20–23 days after the beginning of water treatments. The second sampling was performed on a species-by-species basis: between 35 and 58 days after the start of the water treatments (Supplementary material A).

The following growth variables were measured for all plants sampled on each date: (1) plant height (cm$\bullet$plant^{-1}, with a ruler), (2) leaf area (cm$^2\bullet$plant^{-1}, using a LI-3100 Area Meter; LI-COR Environmental GmbH, Bad Homburg, Germany), (3) root, leaf, stem and, from flowering onwards, reproductive biomass (g$\bullet$plant^{-1}, determined separately by weighing after 48-hour oven dried at 80 °C).

For each plant, ratios of two measured variables enabled to calculate morphological traits of interest: specific leaf area (leaf area to aboveground biomass, SLA in cm$^2\bullet$g^{-1}), height to aboveground biomass ratio (HBR in cm$\bullet$g^{-1}), root to total biomass ratio (RBR in g$\bullet$g^{-1}) and leaf to aboveground biomass ratio (LBR in g$\bullet$g^{-1}).

2.3. Data analysis

2.3.1. Quantifying plant water stress

The response of SLA to mean transpirable fraction of soil water (FTSW) from the beginning of water treatments varied for a given plant species and stage between the current experiment and the previous work of Moreau et al. (2022) (Supplementary material B). Indeed, similar water availability levels cannot necessarily answer to the plants' needs depending on their sizes and stages, thus resulting in different levels of plant stress. For this reason, the morphological traits were analysed in response to a plant water stress index rather than a soil water availability index. The water stress index was based on plant leaf area, which is highly sensitive to water stress (Gómez-del-Campo et al., 2002; Karamanos, 1978). The water stress index was calculated as follows:

(1) the potential value of plant leaf area was estimated by averaging values over the n plants (n=5 plants for all species, except n=4 for ABUTH) exposed to maximum water availability (95 % of field capacity).
(2) the water stress index WSI_{psx} of plant p of species s at stage x (dimensionless) was calculated as follows:

$$WSI_{psx} = \max(0; 1 - \frac{Plant\_leaf\_area_{psx}}{Maximum\_leaf\_area_{sx}}) \quad (1)$$

$Plant\_leaf\_area_{psx}$ (cm) corresponds to the leaf area of plant p of species s at stage x and $Maximum\_leaf\_area_{sx}$ (cm) is the averaged leaf area of plants of species s that grew in optimal water conditions at stage x (95 % of field capacity). Thus, the water stress index is proportional to the reduction of plant leaf area. Given that the ratio of $Plant\_leaf\_area_{psx}$ to $Maximum\_leaf\_area_{sx}$ can exceed 1 for some plants grown in optimal water conditions, their water stress index is set at 0, so that WSI_{psx} ranges from 0 (the plant is not experiencing water stress: its leaf area equals or exceeds the value of $Maximum\_leaf\_area_{sx}$) to 1 (plant water stress is maximal and triggers plant death).

2.3.2. Calculating response parameters

In order to assess the species-inherent response to water stress, four generic two-parameter non-linear regression equations applicable to all species and stages were fitted to the values of each trait vs water stress index for each of the 13 species × stage combinations (6 species × 2 stages, and ALOMY at early vegetative stage). Equations were fitted using either the Nonlinear Least Squares function ('nls', package 'stats', Hastie and Chambers 1992) of R (version 4.2.3) for SLA, whose equation cannot be linearized (Eq. (2)), or the Linear Model function ('lm', package 'stats', Hastie and Chambers 1992) of R for HBR, LBR and RBR, whose equations are linearizable and have the same form (traits grouped in Eq. (3)).

For each species × stage combination, one parameter assessed the potential trait value in unstressed conditions, and the other the species response to water stress. The equations were as follows, with parameters written in ***bold italic***:

For specific leaf area (SLA):

$$SLA_{sx} = \boldsymbol{SLA0_{sx}} \bullet \left(1 - \exp\left(-\boldsymbol{SLA\_resist_{sx}} \bullet \left(1 - WSI_{ps}\right)\right)\right) \tag{2}$$

SLA_{sx} (cm^2·g^{-1}) is the specific leaf area (SLA) of species s at stage x for a given value of water stress index for plant p of species s (WSI_{ps}, dimensionless). $\boldsymbol{SLA0_{sx}}$ (cm^2·g^{-1}) is the SLA for plants of species s that did not undergo water stress at stage x and $\boldsymbol{SLA\_resist_{sx}}$ (dimensionless) reflects the species response of SLA to water stress at stage x.

For height to aboveground biomass ratio (HBR), root to total biomass ratio (RBR) and leaf to aboveground biomass ratio (LBR):

$$\begin{aligned} Trait_{sx} &= \boldsymbol{Trait0_{sx}} \bullet \exp\left(\boldsymbol{Trait\_mu_{sx}} \bullet WSI_{ps}\right) \\ &\Longleftrightarrow \quad \log_n\left(Trait_{sx}\right) \\ &= \log_n\left(\boldsymbol{Trait0_{sx}}\right) + \boldsymbol{Trait\_mu_{sx}} \bullet WSI_{ps} \end{aligned} \tag{3}$$

For each trait (HBR_{sx}, RBR_{sx} and LBR_{sx}), the equation uses two parameters. $\boldsymbol{Trait0_{sx}}$ is the value of the trait taken by plants of species s that did not undergo water stress at stage x (respectively $\boldsymbol{HBR0_{sx}}$ in cm·g^{-1}, $\boldsymbol{RBR0_{sx}}$ in g·g^{-1} and $\boldsymbol{LBR0_{sx}}$ in g·g^{-1}) and $\boldsymbol{Trait\_mu_{sx}}$ (respectively, $\boldsymbol{HBR\_mu_{sx}}$, $\boldsymbol{RBR\_mu_{sx}}$ and $\boldsymbol{LBR\_mu_{sx}}$, dimensionless) is the response of the trait to water stress for species s at stage x. These equations can be linearized after $\log_n$-transformation, whose slopes are the response parameters of HBR, RBR and LBR.

The values of the response parameters ($\boldsymbol{SLA\_resist_{sx}}$, $\boldsymbol{HBR\_mu_{sx}}$, $\boldsymbol{RBR\_mu_{sx}}$ and $\boldsymbol{LBR\_mu_{sx}}$) indicate the effect of increasing water stress on the morphological traits. (1) $\boldsymbol{SLA\_resist_{sx}}$ reflects the resistance of SLA to water stress: the higher the value of the parameter, the stronger the water stress must be to decrease SLA. (2) $\boldsymbol{HBR\_mu_{sx}}$, $\boldsymbol{RBR\_mu_{sx}}$ and $\boldsymbol{LBR\_mu_{sx}}$ reflect the sensitivity of HBR, RBR and LBR to water stress. Traits show greater sensitivity to water stress as the values of their response parameters move away from zero: more positive values correspond to a greater increase in the trait with increasing water stress, while more negative values indicate a greater decrease. These traits have a generic response to water stress if the values of their response parameter have the same sign for all species × stage combinations (when significant, only positive or only negative values).

2.3.3. *Comparing response parameters among species and stages*

After identifying the general trends in the response of morphological traits, we specifically investigated the diversity of trait responses to water stress ($\boldsymbol{SLA\_resist_{sx}}$, $\boldsymbol{HBR\_mu_{sx}}$, $\boldsymbol{RBR\_mu_{sx}}$, $\boldsymbol{LBR\_mu_{sx}}$) according to the species × stage combinations, in two steps.

1. We tested, for each species × stage combination, whether a given trait significantly responded to increasing water stress. For RBR, LBR and HBR, whose regression equations were linearizable, slopes $\boldsymbol{Trait\_mu_{sx}}$ were compared to 0 through 'summary' base function of R. For the SLA with its non-linearizable regression equation, Student tests were used to compare $\boldsymbol{SLA_resist_{sx}}$relatively to 0.
2. We characterised the differences in values of response parameters among species × stage combinations (i.e. diversity in the direction and in the intensity of responses) for each trait. Parameters were compared through two-by-two Student tests ($\boldsymbol{SLA\_resist_{sx}}$) or through slope comparison tests ($\boldsymbol{HBR\_mu_{sx}}$, $\boldsymbol{RBR\_mu_{sx}}$, $\boldsymbol{LBR\_mu_{sx}}$), with function 'emstrends' of 'lsmeans' R package (Lenth, 2016).

2.3.4. *Identifying the main factors affecting response parameters*

To assess the weight of the species effect on morphological traits relatively to other factors (plant stage, water stress intensity), traits were analysed as a function of species, plant stage, water stress as well as all double and triple interactions, through type-III analyses of variance ('Anova' function of 'car' R package, Fox et al. 2023). Partial R^2 were calculated from sum of squares ('rsq.partial' function of 'rsq' R package, Zhang 2023) to determine which factor (species, stage or water stress) explained most of the trait variability.

For a more detailed analysis of the species effect, other analyses of variance were run, where the species effect was replaced by (1) either the status (crop/weed) of the species (2) or the clade (monocotyledons/dicotyledons) of the species.

2.3.5. *Building a typology based on response parameters*

In order to identify traits with similar or contrasting behaviours under water stress, Pearson correlation tests among response parameters of traits were performed ('cor.test' function of 'stats' R package). Categories of morphological behaviours under water stress among species × stage combinations were then built, through a Principal Component Analysis with the response parameters of the four morphological traits (SLA, HBR, LBR and RBR, functions 'PCA' and 'fviz_pca_biplot' of 'FactoMineR' and 'factoextra' R packages, Lê et al. 2008). Individuals (i.e. species × stage combinations) were clustered by using the 'Hierarchical Clustering on Principal Components' (HCPC) function of FactoMineR.

3. Results

3.1. *General response patterns of morphological traits to water stress*

General response patterns of the morphological traits to water stress were highlighted across all species × stage combinations (Fig. 1). Two traits were deeply affected by water stress. (1) Specific leaf area (SLA) (Fig. 1A) decreased exponentially with increasing water stress: plant leaves became smaller and per unit leaf biomass. SLA had the greatest response amplitude, given that leaf area tended towards 0 cm^2 for all species as water stress intensified. (2) Height to aboveground biomass ratio (HBR) (Fig. 1B) was also greatly impacted by water stress: in contrast to SLA which decreased, HBR increased exponentially with increasing water stress. Plants grew taller from a given aboveground biomass when water stress increased. HBR also varied greatly, increasing by up to tenfold (for flowering GERDI) when the stress index reached its maximum value.

Root to total biomass ratio (RBR) (Fig. 1C) and leaf biomass to aboveground biomass ratio (LBR) (Fig. 1D) were less affected, and the response pattern differed between the two traits. (1) RBR increased exponentially with increasing water stress and even at low levels of stress: plants tended to allocate proportionally more biomass to their roots when stressed. RBR varied less than HBR, with a maximum threefold increase (for flowering GERDI) under extreme water stress. (2) For LBR, the direction of the response varied according to the species × stage combinations (either increased or decreased). LBR also was the trait with the lowest response amplitude to water stress (between 15 % decrease and 55 % increase only). Late vegetative wheat was the species × stage combination that the most increased the allocation of aboveground biomass to its stems (vs leaves), whereas flowering GERDI was the species × stage combination that redistributed the lowest proportion of aboveground biomass to its stems and flowers in response to water stress.

3.2. *Morphological responses to water stress vary with species and stages*

Despite the general trends in morphological responses, there was a diversity in the behaviour of species × stage combinations when faced with water stress (Fig. 2). Values of species response parameters of morphological traits were used to quantify this diversity.

Specific leaf area (SLA) (Fig. 2A) and height to aboveground biomass ratio (HBR) (Fig. 2B) were significantly impacted by water stress for 12 out of 13 species × stage combinations, whereas root to total biomass ratio (RBR) (Fig. 2*C*) and leaf to aboveground biomass ratio (LBR) (Fig. 2D) were respectively impacted for respectively 8 and 9 out of 13 combinations.

SLA was the trait whose response parameter $\boldsymbol{SLA\_resist_{sx}}$ took the

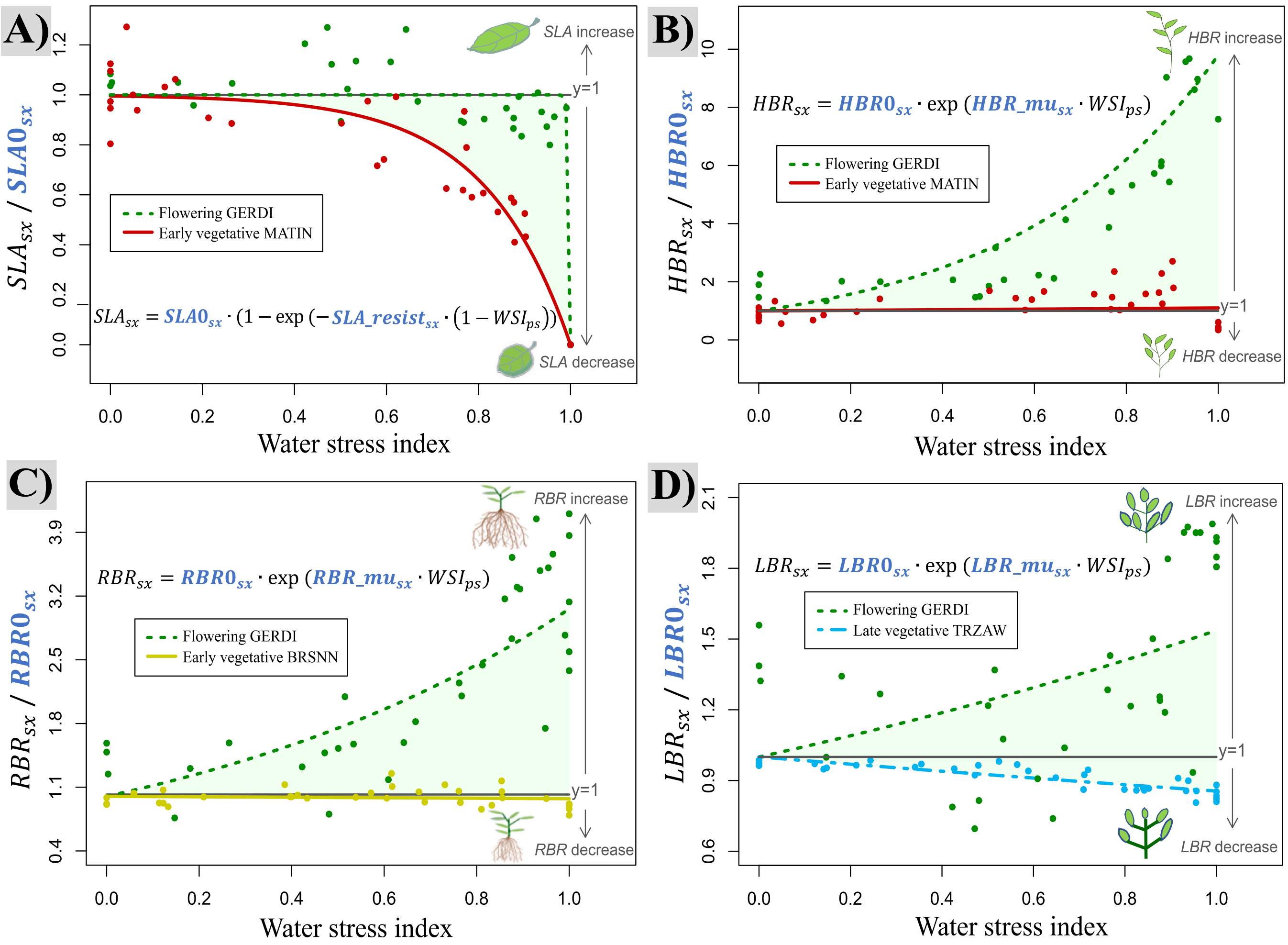

Fig. 1. Changes in morphological traits as a function of water stress index (WSI_{psx}) for two species × stage combinations with contrasted responses. A) Specific leaf area (SLA); B) Height to aboveground biomass ratio (HBR); C) Root biomass to total biomass ratio; D) Leaf biomass to aboveground biomass ratio (LBR). Each data point corresponds to a plant. Generic regression equations are written with parameters in **bold italic blue** (indexes: p = plant; s = species; x = stage). All traits were normalised by the maximum value they reached under unstressed conditions (***SLA0**$_{sx}$*, ***HBR0**$_{sx}$*, ***LBR0**$_{sx}$*, ***RBR0**$_{sx}$*, respectively). Only extreme curves are shown on the graphs, whose colours depend on the species (designated by their EPPO codes) and whose line types depend on the plant stage (early vegetative: continuous curves; late vegetative: non-regular dotted curves; flowering: regular dotted curves). The range of variation of morphological traits among all species × stage combinations is coloured in pale green.

widest range of values (all positive values) among species × stage combinations, with 7 different groups. The lower the response parameter, the more sensitive the SLA was to water stress, i.e. leaves became smaller per unit leaf biomass even at low stress levels. The highest values were taken by dicotyledonous species (GERDI, ABUTH and BRSNN, groups d to f), especially at late stages (groups f and g), which kept their usual SLA until extreme levels of stress. For all species, SLA was more sensitive to water stress at the early vegetative vs later stages.

For height to aboveground biomass ratio (HBR), the values taken by the response parameter ***HBR_mu**$_{sx}$* were divided into 4 groups. They ranged from species × stage combinations that responded the most, i.e. species at late vegetative or flowering stages that grew the most per unit aboveground biomass in response to water stress (group a, highest values of ***HBR_mu**$_{sx}$*), to combinations whose ***HBR_mu**$_{sx}$* were low or even not significantly different from zero (groups c and d, little growth increase per unit aboveground biomass). Those two groups included all species at early vegetative stages.

As for ***HBR_mu**$_{sx}$*, the response parameter of root to total biomass ratio (***RBR_mu**$_{sx}$*) was divided into 4 groups. Combinations involving GERDI and ABUTH (two dicotyledonous weeds, groups a and b) took the highest values of ***RBR_mu**$_{sx}$*, especially at the flowering stage, and greatly increased biomass allocation to roots in response to water stress. Other species responded less, whatever their stages, clades or status.

Leaf to aboveground biomass ratio (LBR) was the trait whose values of the response parameter (***LBR_mu**$_{sx}$*) varied the least, with only three unbalanced groups. Species × stage combinations with the highest positive values of ***LBR_mu**$_{sx}$* increased aboveground biomass allocation to leaves to the detriment of stems and reproductive organs in response to water stress (groups a and b). They were dicotyledonous species (GERDI, ABUTH, MATIN) at the flowering stage. The LBR of the other species × stage combinations (group c) either did not vary (values not significantly different from zero) or decreased (negative values) in response to water stress, i.e. they attributed less biomass to leaves than in unstressed conditions. All monocotyledonous species and the two crop species tended to attribute more biomass to their stems (vs leaves).

3.3. Drivers of the diversity in species × stage responses to water stress

When analysing morphological traits as a function of experimental factors, the species factor alone explained the main part of the variability for all traits (partial R^2 ranging from 0.30 to 0.59), followed by water stress index for the two size-related traits (SLA and HBR) or plant stage for the two biomass-allocation traits (LBR and RBR) (Table 2). The effects of (1) plant species and (2) plant stage on trait response to water

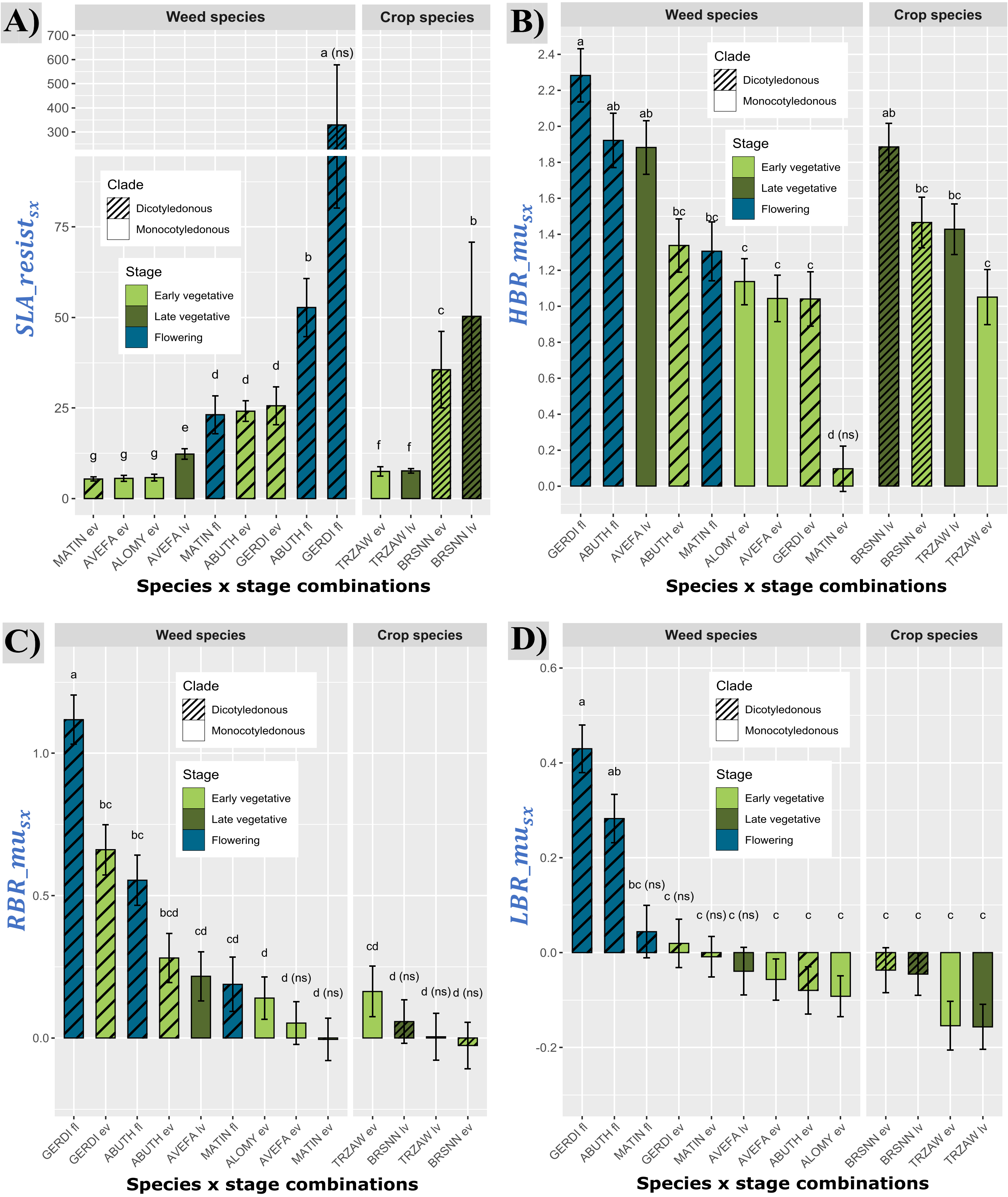

Fig. 2. Diversity in values of response parameters of morphological traits to water stress, according to species × stage combinations. A) Specific leaf area to water stress (***SLA_resist***$_{sx}$); B) height to aboveground biomass ratio to water stress (***HBR_mu***$_{sx}$); C) Root to total biomass ratio to water stress (***RBR_mu***$_{sx}$); D) Leaf to aboveground biomass ratio to water stress (***LBR_mu***$_{sx}$). Error bars represent SE of parameter values, n = 7 treatments × 5 repetitions (only 4 repetitions for ABUTH). Significance letters distinguish responses of associated morphological traits according to the species × stage combinations (***SLA_resist***$_{sx}$: two-by-two Student tests; ***HBR_mu***$_{sx}$, ***LBR_mu***$_{sx}$ and ***RBR_mu***$_{sx}$: two-by-two slope comparisons, R 'pairs' function of 'lsmeans' package). Non-significant (ns) values of response parameters at $p < 0.05$ are given in brackets.

Table 2
Partial R^2 values of models representing each morphological trait as a function of species, stage and water stress.

Factor	Specific leaf area SLA[1]	Height Biomass Ratio $\log_n$(HBR)	Leaf Biomass Ratio $\log_n$(LBR)	Root Biomass Ratio $\log_n$(RBR)
Species	0.30	0.51	0.59	0.41
Plant stage	0.01	0.13	0.16	0.10
Water_stress	0.28	0.23	ns	0.07
Species:Stage	0.01	0.01	0.05	0.04
Species: Water_stress	0.06	0.02	0.01	0.07
Stage: Water_stress	ns	0.02	0.02	0.01
Species:Stage: Water_stress	ns	ns	0.01	ns
Total variance explained	**0.66**	**0.92**	**0.84**	**0.70**

All morphological traits were $\log_n$-transformed ([1]except specific leaf area (SLA), whose applied regression equation was non-linearizable). All values were significant at $p < 0.05$, otherwise the factor was defined as not-significant ('ns').

stress were significant (except plant stage for SLA response to water stress), but did not account for much of the overall variability (partial R^2 ranging from 0.01 to 0.07 for species × water stress interaction and from 0 to 0.02 for stage × water stress interaction).

The differential morphological responses to water stress among plant species were more precisely analysed, to search for generic rules and to determine whether they could be related to the effects of (1) the species clade (monocotyledonous/dicotyledonous) and/or (2) status (weed/crop) (Table 3). None of these two factors significantly explained the variations, despite a close-significant relationship for clade-dependent LBR response to water stress. There was an overall LBR decrease with increasing water stress in monocotyledons, versus all possibilities (increase, no effect or decrease) in dicotyledons.

3.4. *Typology of responses to water stress among species × stage combinations*

In order to identify similarities among species adaptation to water stress, Pearson correlations between parameters of morphological responses were analysed. There were strong positive correlations between LBR and RBR response parameters (Pearson correlation coefficient $r = 0.83$, $p < 0.01$), RBR and SLA response parameters ($r = 0.81$, $p < 0.01$) and LBR and SLA response parameters ($r = 0.82$, $p < 0.01$). Thus, species × stage combinations that the most increased the proportion of biomass allocated to their roots in response to water stress (high **RBR_mu_{sx}**) were also those that the most increased the proportion of aboveground biomass allocated to their leaves (high **LBR_mu_{sx}**) and whose leaves shrank per unit leaf biomass only when water stress became extreme (high **SLA_resist_{sx}**). Another significant positive correlation was observed between HBR and SLA response parameters ($r = 0.59$, $p = 0.03$). Species × stage combinations that grew taller per unit aboveground biomass in response to water stress only shrank their leaves per unit leaf biomass at extreme levels of stress.

Table 3
Effect of species clade (mono/dicotyledonous) and status (crop/weed) on the parameters describing morphological response to water stress.

Factor	SLA_resist_{sx}	LBR_mu_{sx}	RBR_mu_{sx}	HBR_mu_{sx}
Status				
Weed	non-significant at $p < 0.1$	non-significant at $p < 0.1$	non-significant at $p < 0.1$	non-significant at $p < 0.1$
Crop				
Partial R^2				
p-value				
Clade				
Monocotyledonous	non-significant at $p < 0.1$	-0.10	non-significant at $p < 0.1$	non-significant at $p < 0.1$
Dicotyledonous		0.08		
Partial R^2		0.28		
p-value		0.06		

For each trait × factor combination, mean values of the response parameters were determined by factor levels, and type-III ANOVA were performed (linear models of response parameters as a function of factors).

A typology of responses to water stress was highlighted through a Principal Component Analysis. Six behaviours were identified, with heterogeneous cluster sizes (Fig. 3).

Seven species × stage combinations behaved similarly: ABUTH, ALOMY, AVEFA and BRSNN at early vegetative stage, TRZAW at early and late vegetative stages and MATIN at flowering stage (cluster 1). In response to water stress, they tended to (1) shrink their leaves per unit leaf biomass even at low levels of water stress (low **SLA_resist_{sx}**), (2) expand in height for a given aboveground biomass (high **HBR_mu_{sx}**), (3) reasonably increase the proportion of biomass allocated to the roots (intermediate **RBR_mu_{sx}**) and (4) very slightly reduce the proportion of aboveground biomass allocated to the leaves (low **LBR_mu_{sx}**).

The six other species × stage combinations had diverse morphological responses to water stress. For flowering ABUTH and GERDI (clusters 2 and 3), leaves decreased their area per unit biomass only at extremely high levels of water stress. These combinations were those that (1) increased the proportion of biomass allocated to the roots (vs shoots) and leaves (vs stems and reproductive organs) the most and (2) grew taller for a given aboveground biomass in response to water stress, especially for GERDI.

Early vegetative GERDI (cluster 4) behaved in the same way as the species of cluster 1, but allocated a higher proportion of biomass to roots in response to water stress (comparable to clusters 2 and 3). Morphological responses of rapeseed (BRSNN) and AVEFA at late vegetative stage (cluster 5) were also similar to species × stage combinations of cluster 1, but these combinations tended to grow taller from a given aboveground biomass in response to water stress. Early vegetative MATIN (cluster 6) was the only species × stage combination whose behaviour contradicted the correlations observed between morphological parameters. Although their leaves were smaller per unit leaf biomass, even at low levels of stress, early vegetative MATIN plants did not change the allocation of biomass neither to their roots nor to their leaves, and no change in height was observed for a given aboveground biomass.

4. Discussion

This innovative study is to our knowledge the first to characterise the responses of four morphological traits to water stress (1) on such a large panel of five weed species and two crop species, whereas most past studies focused on one to three species (Chahal et al., 2018; Monaco et al., 2005; Moreau et al., 2022; Noda et al., 2004; Zhou et al., 2020); (2) at both vegetative and flowering stages, where most studies focused on germination and emergence (Singh et al., 2022); (3) and quantitatively across a continuous gradient of seven levels of water availability, and not qualitatively with classes of water availability, i.e. low vs high water availability (Monaco et al., 2005). As far as we know, this is the first work on weed morphological response to water stress that uses response parameters of regression curves as quantifiers of interspecies diversity. This study identified generic response patterns of morphological traits to water stress and also performed comparative ecology to better understand interspecies differences at different phenological stages.

4.1. *Response patterns of morphological traits to water stress are consistent with existing literature and hypotheses on plant functioning*

All four monitored morphological traits responded to water stress. We found that specific leaf area (SLA) strongly decreased in response to water stress, while height to aboveground biomass ratio (HBR) sharply

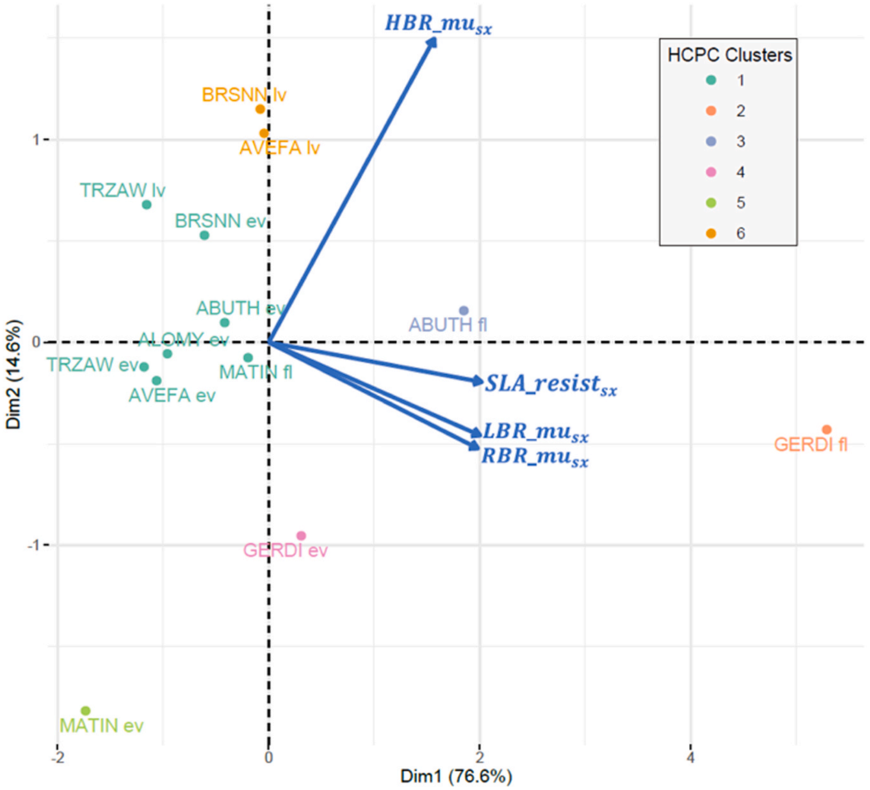

Fig. 3. Hierarchical clustering on principal components (HCPC) of species × stage combinations, from a principal component analysis (PCA) on response parameters of morphological traits to water stress. Response parameters of the four morphological traits to water stress are the four variables of the PCA (**SLA_resist_{sx}** for specific leaf area, **LBR_mu_{sx}** for leaf to aboveground biomass ratio, **RBR_mu_{sx}** root to total biomass ratio, **HBR_mu_{sx}** height to aboveground biomass ratio). Six types of responses were highlighted.

increased with water stress. The response of SLA is consistent with the literature: water stress decreases leaf area per unit leaf biomass, reduces transpiration, and thus plant water demand for photosynthesis (Monaco et al., 2005; Poorter et al., 2009; Zhou et al., 2020). To our knowledge, only the work of Moreau et al. (2022) has investigated the response of HBR to water availability on few species, with similar findings showing that water stress reduced aboveground biomass more than plant height.

The root to total biomass ratio (RBR) was impacted by water stress for most species × stage combinations (8 out of 13). When affected, it always increased in response to water stress. These results are consistent with the functional balance theory (Brouwer, 1962): the plant allocates more biomass to the compartment that takes up the most limiting resource.

No generic trends were observed for the ratio of leaf to aboveground biomass (LBR), which either decreased (7 cases out of 13), remained unchanged (4 cases out of 13) or increased (2 cases out of 13) in response to water stress. These results are partially consistent with the literature, where LBR is either increased (Chahal et al., 2018) or not impacted (Lu et al., 2014; De Oliveira et al., 2018) in response to water stress.

4.2. New knowledge on plant functioning and comparative ecology

Our results showed a wide range of morphological responses to water stress among species × stage combinations. Most combinations (7 out of 13) experienced smaller leaves per unit leaf area, taller plants per unit leaf aboveground biomass, priority biomass allocation to roots (vs shoots) and a slight reduction of aboveground biomass allocation to leaves (vs stems and reproductive organs). In other words, these cases reduced transpiration and improved water uptake.

Morphological responses of the six other species × stage combinations differed either in the direction of the response (increase, no impact or decrease), especially for biomass-allocation traits RBR and LBR, or in the intensity of the response (same direction but more or less strong changes), particularly for size-based traits SLA and HBR. Correlations between response parameters of traits showed that species × stage combinations that did not decrease their SLA until reaching high levels of stress were also those that increased the most their RBR and LBR. Thus, these combinations favoured one of the mechanisms for adapting to water stress. As defined by Basu et al. (2016), early vegetative GERDI, flowering ABUTH and flowering GERDI were 'water spenders', as they increased water uptake without necessarily reducing demand at low stress levels, whereas early vegetative MATIN were 'water savers', as they reduced demand at low stress levels but did not increase water uptake. Late vegetative rapeseed and AVEFA were theoretically less adapted to water stress, but they kept a better light-interception potential (high HBR).

For all traits, a coupled effect of species and plant stage on morphological response to water stress was highlighted, but differences were mainly explained by the species. Actually, the effects have often been studied separately: on the one hand, the genetic characteristics of species (and cultivars) contribute to explaining the diversity of plant responses to water stress (De Leonardis et al., 2007, 2012). On the other hand, several studies have shown that annual plants are more sensitive to water stress during later stages, particularly reproduction (Nemeskéri et al., 2019; Nemeskéri and Helyes, 2019). The latter report is consistent with our results, as HBR, LBR and RBR tended to be more impacted when plants aged. Only SLA was more impacted at early vegetative stage for all species. As suggested by Poorter et al. (2012), allocation ratios change are less visible at early plant stages in order to maintain light interception ability and facilitate normal growth recovery after the stress event.

To provide genericity beyond the studied species, we tried to identify the determinants of interspecies variability in morphological responses to water stress. Species-dependent responses were neither explained by clade (monocotyledons/dicotyledons) nor status (crop/weed) for SLA, HBR and RBR. There was yet a close-significant effect of clade on LBR response to water stress, with decreased aboveground biomass allocation to leaves for monocotyledons. To our knowledge, this effect has never been highlighted in previous studies. To go further, additional weed/crop and mono/dicotyledonous species need to be tested.

4.3. Implications for weed management

Most studies on weeds mainly focus on the consequences of a given weed flora on the final yield of the crop or its components, due to competition for water and other resources (Maxwell and O'Donovan, 2007; Rejmánek et al., 1989), without discriminating individual effects of light, water or nitrogen competition. Our work complements these studies, in that it mimicked competition for water only, on the assumption that the plant reacts in the same way whether the water stress is caused by uptake from neighbouring plants, drought, or both simultaneously.

On top of water stress, plants can also be confronted with other stresses (shading and nitrogen limitation) during their life cycle. In order to understand the overall behaviour of plants, it is therefore essential to study the effects of water stress on plant morphology in conjunction with those of the other major stresses (Table 4).

4.3.1. Water vs nitrogen stress

plant responses are similar for three out of four traits, with a difference for height to aboveground biomass ratio (HBR). In response to water and nitrogen stress, specific leaf area (SLA) decreases, leading and smaller leaves per unit leaf biomass. This allows, on one hand, reducing plant water demand for leaf transpiration (Chahal et al., 2018; Monaco et al., 2005) and, on the other hand, optimising nitrogen use (Freschet et al., 2015). Plants also allocate more biomass to their roots (vs aboveground organs) under both nitrogen and water stress to enhance soil exploration and look for new resource pools (Brouwer, 1962; Carretero et al., 2014). However, while nitrogen stress has a very slight effect on HBR and in both directions (Perthame et al., 2022), water stress leads to a sharp increase in HBR. A higher HBR does not necessarily lead to a physiological advantage for plants, but it can be interpreted as a greater impact of water stress on aboveground biomass than on height (Moreau et al., 2022).

Table 4
Responses of morphological traits to water stress, nitrogen stress and shading.

Type of stress	**Morphological trait**			
	Specific leaf area (SLA)	Height to aboveground biomass ratio (HBR)	Leaf biomass to aboveground biomass ratio (LBR)	Root biomass to total biomass ratio (RBR)
Water stress (this article)	⬇	⬆	↑ ↓	⬆
Nitrogen stress (Perthame et al., 2022, 2020; Pointurier et al., 2021)	⬇	↑ ↓	↑ ↓	⬆
Light stress (Colbach et al., 2020; Munier-Jolain et al., 2014; Pointurier et al., 2021)	⬆	⬆	↑ ↓	**0**

Stresses can lead to an increase (⬆), a decrease (⬇) or have no effect (**0**) on morphological traits, depending on the species × stage combinations. For each type of stress, the larger the arrows, the greater the effect of the stress on the morphological trait.

4.3.2. Water stress vs shade

plant responses are similar for only two variables (LBR and HBR) and they differ for SLA and RBR. Under water stress and shade, leaf to aboveground biomass ratio can either increase, decrease or remain unchanged according to species and stages. Moreover, both stresses trigger a sharp increase in HBR. For shaded plants, HBR increase is mainly driven by height increase and stem elongation (etiolation), which provide a competitive advantage to get access to light (Colbach et al., 2021; Collins and Wein, 2000; Munier-Jolain et al., 2014). However, unlike under water stress, shaded plants increase their SLA to maximise light interception and carbon gain per unit leaf biomass (Liu et al., 2016) and they do not change their RBR (Pointurier et al., 2021).

Such comparisons of the effects of a limitation in several resources, particularly on weed species, had never been studied before. Given that the most limiting resources to plant growth generally vary during their life cycle, it is nearly impossible to predict their morphological evolution. Depending on the cropping systems (crops and cultivars, rotations, technical operations such as fertilisation, ploughing or irrigation), the pedoclimates and the weed seedbanks, one or more resources may be insufficient to meet the needs of all plants at several stages of the growing cycle and crop/weed interactions can be highly variable (Little et al., 2021). Moreover, the morphological responses of plants to stresses are not the only factors that determine a plant's level of competitiveness, which also depends on germination, emergence, photosynthesis or initial growth rate for example (Travlos et al., 2011). Emergence timing of weeds vs crops is actually more important for yield or seed production than their density or proximity, indicating that emergence timing trumps competitive ability (Fahad et al., 2015).

Data such as that produced in this article is crucial for feeding and improving mechanistic models, to understand the relative impact of biological phenomena linked to weed/crop interactions on crop growth and yield, as well as on weed flora dynamics (competition for light, competition for water, competition for nutrients, allelopathy, etc.) (Belz, 2007; Colbach et al., 2021). Mechanistic models represent numerous biophysical processes and cropping techniques interacting with the soil and the climate (Pointurier et al., 2021). Some of them, such as FLORSYS, are also individual-based: they simulate the growth of each plant (crop or weed) in a virtual field (Colbach et al., 2021). This makes it possible to represent the competitive phenomena among dozens of weed and crop species with different properties, and to predict the outcomes of crop/weed interactions.

5. Conclusion

This original study, both in its objective and approach, allowed finely characterising the diversity in morphological responses to water stress within a panel of five weed species and two crop species. Generic trends were identified for responses of specific leaf area (SLA decrease), height to aboveground biomass ratio and root to total biomass ratio (HBR and RBR increase) to water stress, but not for leaf to aboveground biomass ratio (LBR), with either increase or decrease. Despite generic trends to reduce transpiration and increase water uptake, weed and crop responses to water stress were diverse and both species- and stage-dependent, clustered in six unbalanced categories of behaviours. This work also highlighted a close-significant relationship for clade-dependent LBR response to water stress.

By providing key insights into weed morphological plasticity, this work complements previous studies that have investigated the impacts of both nitrogen stress and shade on the morphology of several crops and weeds. Additional research is needed to (1) characterise the morphological response to water stress of more species and (2) go even further in the identification of factors that explain the interspecies diversity in morphological trait responses to water stress, in order to predict the behaviour of a wider range of weed species.

CRediT authorship contribution statement

Quentin Cournault: Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Nathalie Colbach:** Writing – review & editing, Supervision, Resources, Methodology, Funding acquisition, Conceptualization. **Hugues Busset:** Resources, Methodology. **Annick Matejicek:** Resources, Methodology. **Pauline Souche-Suchovsky:** Resources, Methodology, Data curation. **Marion Prudent:** Writing – review & editing, Resources. **Delphine Moreau:** Writing – review & editing, Supervision, Resources, Methodology, Funding acquisition, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

This project was supported by INRAE, the French Ministry of Agriculture (Ministère de l'Agriculture et de la Souveraineté Alimentaire) which funded the Ph.D. thesis of Quentin Cournault, the COPRAA project funded by the Office Français de la Biodiversité (OFB), as part of an Ecophyto call launched by the French Ministries in charge of Ecology, Agriculture, Health and Research, the PPR projects SPECIFICS (ANR-20-PCPA-0008) and MOBIDIV (ANR-20-PCPA-0006) funded by the 'Growing and Protecting crops Differently' French Priority Research Program (PPR-CPA), part of the national investment plan operated by the French National Research Agency (ANR), as well as the Horizon Europe project IntercropVALUES. The authors thank the staff of the 'serres_4PMI' platform of INRAE Dijon for technical assistance during the greenhouse experiment.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.envexpbot.2024.105825.

References

Basu, S., Ramegowda, V., Kumar, A., Pereira, A., 2016. Plant adaptation to drought stress. F1000Res 5. https://doi.org/10.12688/f1000research.7678.1.

Belz, R.G., 2007. Allelopathy in crop/weed interactions — an update. Pest Manag. Sci. 63, 308–326. https://doi.org/10.1002/ps.1320.

Brouwer, R., 1962. Nutritive influences on the distribution of dry matter in the plant. Neth. J. Agric. Sci. 10, 399–408. https://doi.org/10.18174/njas.v10i5.17581.

Carretero, R., Bert, F.E., Podestá, G., 2014. Maize root architecture and water stress tolerance: an approximation from crop models. Agron. J. 106, 2287–2295. https://doi.org/10.2134/agronj14.0214.

Chahal, P.S., Irmak, S., Jugulam, M., Jhala, A.J., 2018. Evaluating effect of degree of water stress on growth and fecundity of palmer amaranth (*Amaranthus palmeri*) using soil moisture sensors. Weed Sci. 66, 738–745. https://doi.org/10.1017/wsc.2018.47.

Colbach, N., Colas, F., Cordeau, S., Maillot, T., Queyrel, W., Villerd, J., Moreau, D., 2021. The FLORSYS crop-weed canopy model, a tool to investigate and promote agroecological weed management. Field Crops Res. 261, 108006 https://doi.org/10.1016/j.fcr.2020.108006.

Colbach, N., Gardarin, A., Moreau, D., 2019. The response of weed and crop species to shading: which parameters explain weed impacts on crop production? Field Crops Res 238, 45–55. https://doi.org/10.1016/j.fcr.2019.04.008.

Colbach, N., Munier-Jolain, N., Dugué, F., Gardarin, A., Strbik, F., Moreau, D., 2020. The response of weed and crop species to shading. How to predict their morphology and plasticity from species traits and ecological indexes? Eur. J. Agron. 121, 126158 https://doi.org/10.1016/j.eja.2020.126158.

Collins, B., Wein, G., 2000. Stem elongation response to neighbour shade in sprawling and upright *Polygonum* species. Ann. Bot. 86, 739–744. https://doi.org/10.1006/anbo.2000.1233.

De Leonardis, A.M., Marone, D., Mazzucotelli, E., Neffar, F., Rizza, F., Di Fonzo, N., Cattivelli, L., Mastrangelo, A.M., 2007. Durum wheat genes up-regulated in the early phases of cold stress are modulated by drought in a developmental and genotype dependent manner. Plant Sci. 172, 1005–1016. https://doi.org/10.1016/j.plantsci.2007.02.002.

De Leonardis, A.M., Petrarulo, M., Vita, P.D., Mastrangelo, A.M., 2012. Genetic and molecular aspects of plant response to drought in annual crop species. in: Advances in Selected Plant Physiology aspects. IntechOpen. https://doi.org/10.5772/31352.

De Oliveira, F.S., Gama, D.R., da, S., Dombroski, J.L.D., Silva, D.V., Filho, F., de, S.O., Neta, T.R., De Souza, M.M., 2018. Competition between cowpea and weeds for water: effect on plants growth. Rev. Bras. Cienc. Agrar 13, 1–7. https://doi.org/10.5039/agraria.v13i1a5507.

Fahad, S., Hussain, S., Chauhan, B.S., Saud, S., Wu, C., Hassan, S., Tanveer, M., Jan, A., Huang, J., 2015. Weed growth and crop yield loss in wheat as influenced by row spacing and weed emergence times. Crop Prot. 71, 101–108. https://doi.org/10.1016/j.cropro.2015.02.005.

Fox, J., Weisberg, S., Price, B., 2023. car: companion to applied regression. R. Package Version 3 1–2. ⟨https://CRAN.R-project.org/package=car⟩.

Freschet, G.T., Swart, E.M., Cornelissen, J.H.C., 2015. Integrated plant phenotypic responses to contrasting above- and below-ground resources: key roles of specific leaf area and root mass fraction. N. Phytol. 206, 1247–1260. https://doi.org/10.1111/nph.13352.

Gardarin, A., Guillemin, J.-P., Munier-Jolain, N.M., Colbach, N., 2010. Estimation of key parameters for weed population dynamics models: base temperature and base water potential for germination. Eur. J. Agron. 32, 162–168. https://doi.org/10.1016/j.eja.2009.09.006.

Gleick, P.H., 1989. Climate change, hydrology, and water resources. Rev. Geophys. 27, 329–344. https://doi.org/10.1029/RG027i003p00329.

Gómez-del-Campo, M., Ruiz, C., Lissarrague, J.R., 2002. Effect of water stress on leaf area development, photosynthesis, and productivity in Chardonnay and Airén grapevines. Am. J. Enol. Vitic. 53, 138–143. https://doi.org/10.5344/ajev.2002.53.2.138.

Hastie, T.J., Chambers, J.M., 1992. Statistical Models in S, Wadsworth & Brooks/Cole Computer Science Series. Wadsworth & Brooks/Cole, Pacific Grove.

Hill, M.O., Mountford, J.O., Roy, D.B., Bunce, R.G.H., 1999. Ellenberg's indicator values for British plants. ECOFACT Volume 2 Technical Annex. Institute of Terrestrial Ecology, Huntingdon.

Jeudy, C., Adrian, M., Baussard, C., Bernard, C., Bernaud, E., Bourion, V., Busset, H., Cabrera-Bosquet, L., Cointault, F., Han, S., Lamboeuf, M., Moreau, D., Pivato, B., Prudent, M., Trouvelot, S., Truong, H.N., Vernoud, V., Voisin, A.-S., Wipf, D., Salon, C., 2016. RhizoTubes as a new tool for high throughput imaging of plant root development and architecture: test, comparison with pot grown plants and validation. Plant Methods 12, 31. https://doi.org/10.1186/s13007-016-0131-9.

Karamanos, A.J., 1978. Water stress and leaf growth of field beans (*Vicia faba* L.) in the field: leaf number and total leaf area. Ann. Bot. 42, 1393–1402. https://doi.org/10.1093/oxfordjournals.aob.a085586.

Lê, S., Josse, J., Husson, F., 2008. FactoMineR: an R package for multivariate analysis. J. Stat. Softw. 25, 1–18. https://doi.org/10.18637/jss.v025.i01.

Lenth, R.V., 2016. Least-squares means: the R package lsmeans. J. Stat. Softw. 69, 1–33. https://doi.org/10.18637/jss.v069.i01.

Li, D., Quan, C., Song, Z., Li, X., Yu, G., Li, C., Muhammad, A., 2021. High-Throughput Plant Phenotyping Platform (HT3P) as a novel tool for estimating agronomic traits from the lab to the field. Front. Bioeng. Biotechnol. 8.

Little, N.G., DiTommaso, A., Westbrook, A.S., Ketterings, Q.M., Mohler, C.L., 2021. Effects of fertility amendments on weed growth and weed–crop competition: a review. Weed Sci. 69, 132–146. https://doi.org/10.1017/wsc.2021.1.

Liu, Y., Dawson, W., Prati, D., Haeuser, E., Feng, Y., van Kleunen, M., 2016. Does greater specific leaf area plasticity help plants to maintain a high performance when shaded? Ann. Bot. 118, 1329–1336. https://doi.org/10.1093/aob/mcw180.

Lu, Y.J., Wang, M., Ge, Y., Fu, C.-X., Chang, J., 2014. Response of photosynthetic and growth characteristic of *Mosla chinensis* and congenerous weed *M. scabra* to soil water content. Russ. J. Ecol. 45, 367–374. https://doi.org/10.1134/S106741361405018X.

Ma, D., Carpenter, N., Amatya, S., Maki, H., Wang, L., Zhang, L., Neeno, S., Tuinstra, M. R., Jin, J., 2019. Removal of greenhouse microclimate heterogeneity with conveyor system for indoor phenotyping. Comput. Electron. Agric. 166, 104979 https://doi.org/10.1016/j.compag.2019.104979.

Maxwell, B.D., O'Donovan, J.T., 2007. Understanding weed-crop interactions to manage weed problems. Non-cHemical Weed Management: Principles, Concepts and Technology. CABI Books, pp. 17–33. https://doi.org/10.1079/9781845932909.0017.

Monaco, T.A., Johnson, D.A., Creech, J.E., 2005. Morphological and physiological responses of the invasive weed Isatis tinctoria to contrasting light, soil-nitrogen and water. Weed Res. 45, 460–466. https://doi.org/10.1111/j.1365-3180.2005.00480.x.

Moreau, D., Abiven, F., Busset, H., Matejicek, A., Pagès, L., 2017. Effects of species and soil-nitrogen availability on root system architecture traits – study on a set of weed and crop species. Ann. Appl. Bot. 171, 103–116. https://doi.org/10.1111/aab.12355.

Moreau, D., Busset, H., Matejicek, A., Prudent, M., Colbach, N., 2022. Water limitation affects weed competitive ability for light. A demonstration using a model-based approach combined with an automated watering platform. Weed Res. 62, 381–392. https://doi.org/10.1111/wre.12554.

Munier-Jolain, N.M., Collard, A., Busset, H., Guyot, S.H.M., Colbach, N., 2014. Investigating and modelling the morphological plasticity of weeds. Field Crops Res. 155, 90–98. https://doi.org/10.1016/j.fcr.2013.09.018.

Nemeskéri, E., Helyes, L., 2019. Physiological responses of selected vegetable crop species to water stress. MDPI Agron. 9, 447. https://doi.org/10.3390/agronomy9080447.

Nemeskéri, E., Molnár, K., Rácz, C., Dobos, A.C., Helyes, L., 2019. Effect of water supply on spectral traits and their relationship with the productivity of sweet corns. MDPI Agron. 9, 63. https://doi.org/10.3390/agronomy9020063.

Noda, H., Muraoka, H., Washitani, I., 2004. Morphological and physiological acclimation responses to contrasting light and water regimes in *Primula sieboldii*. Ecol. Res. 19, 331–340. https://doi.org/10.1111/j.1440-1703.2004.00642.x.

Oerke, E.-C., 2006. Crop losses to pests. J. Agric. Sci. 144, 31–43. https://doi.org/10.1017/S0021859605005708.

Perthame, L., Colbach, N., Brunel-Muguet, S., Busset, H., Lilley, J.M., Matejicek, A., Moreau, D., 2020. Quantifying the nitrogen demand of individual plants in heterogeneous canopies: a case study with crop and weed species. Eur. J. Agron. 119, 126102 https://doi.org/10.1016/j.eja.2020.126102.

Perthame, L., Colbach, N., Busset, H., Matejicek, A., Moreau, D., 2022. Morphological response of weed and crop species to nitrogen stress in interaction with shading. Weed Res. 62, 160–171. https://doi.org/10.1111/wre.12524.

Pointurier, O., Moreau, D., Pagès, L., Caneill, J., Colbach, N., 2021. Individual-based 3D modelling of root systems in heterogeneous plant canopies at the multiannual scale. Case study with a weed dynamics model. Ecol. Model. 440, 109376 https://doi.org/10.1016/j.ecolmodel.2020.109376.

Poorter, H., Niinemets, Ü., Poorter, L., Wright, I.J., Villar, R., 2009. Causes and consequences of variation in leaf mass per area (LMA): a meta-analysis. N. Phytol. 182, 565–588. https://doi.org/10.1111/j.1469-8137.2009.02830.x.

Poorter, H., Niklas, K.J., Reich, P.B., Oleksyn, J., Poot, P., Mommer, L., 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. N. Phytol. 193, 30–50. https://doi.org/10.1111/j.1469-8137.2011.03952.x.

Ramesh, K., Matloob, A., Aslam, F., Florentine, S.K., Chauhan, B.S., 2017. Weeds in a changing climate: vulnerabilities, consequences, and implications for future weed management. Front. Plant Sci. 8, 12. https://doi.org/10.3389/fpls.2017.00095.

Ramos, M.C., Martínez-Casasnovas, J.A., 2014. Soil water variability and its influence on transpirable soil water fraction with two grape varieties under different rainfall regimes. Agric. Ecosyst. Environ. 185, 253–262. https://doi.org/10.1016/j.agee.2013.12.025.

Rejmánek, M., Robinson, G.R., Rejmánková, E., 1989. Weed-crop competition: experimental designs and models for data analysis. Weed Sci. 37, 276–284. https://doi.org/10.1017/S0043174500071903.

Singh, M., Thapa, R., Kukal, M.S., Irmak, S., Mirsky, S., Jhala, A.J., 2022. Effect of water stress on weed germination, growth characteristics, and seed production: a global meta-analysis. Weed Sci. 70, 621–640. https://doi.org/10.1017/wsc.2022.59.

Tardieu, F., 2003. Virtual plants: modelling as a tool for the genomics of tolerance to water deficit. Trends Plant Sci. 8, 9–14. https://doi.org/10.1016/S1360-1385(02)00008-0.

Travlos, I.S., Economou, G., Kanatas, P.J., 2011. Corn and barnyardgrass competition as influenced by relative time of weed emergence and corn hybrid. Agron. J. 103, 1–6. https://doi.org/10.2134/agronj2010.0245.

Zhang, D., 2023. rsq: R-squared and related measures. R package version 2.6. ⟨https://CRAN.R-project.org/package=rsq⟩.

Zhou, H., Zhou, G., He, Q., Zhou, L., Ji, Y., Zhou, M., 2020. Environmental explanation of maize specific leaf area under varying water stress regimes. Environ. Exp. Bot. 171, 103932 https://doi.org/10.1016/j.envexpbot.2019.103932.