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RESEARCH ARTICLE

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# The PhenObs initiative: A standardised protocol for monitoring phenological responses to climate change using herbaceous plant species in botanical gardens

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## Abstract

1. Changes in phenology induced by climate change occur across the globe with important implications for ecosystem functioning and services, species performance and trophic interactions. Much of the work on phenology, especially leaf out and flowering, has been conducted on woody plant species. Less is known about the responses in phenology of herbaceous species induced by global change even though they represent a large and important part of biodiversity worldwide. A globally coordinated research effort is needed to understand the drivers and implications of such changes and to predict effects of global change on plant species phenology and related ecosystem processes.
2. Here, we present the rationale of the PhenObs initiative—botanical gardens as a global phenological observation network. The initiative aims to collect data on plant phenology in botanical gardens which will be used alongside information on plant traits and site conditions to answer questions related to the consequences of global change:
   - A What is the variation in plant phenology in herbaceous species across the growing season and in response to changes in climate?
   - B How can plant phenology be predicted from species' trait composition, provenance, position and extent of the distribution range and species' phylogeny?
   - C What are the implications of this variation with respect to species performance and assembly, biotic interactions (e.g. plant–pollinator interactions) as well as ecosystem processes and services under changing land use and climate?

3. Here, we lay out the development of a straightforward protocol that is appropriate for monitoring phenology across a vast diversity of growth forms of herbaceous species from various habitats and geographical regions.
4. To focus on a key number of stages necessary to capture all aspects of plant species phenology, we analysed associations between 14 phenological stages. These data were derived from a 2-year study on 199 species in four German botanical gardens.
5. Based on the relationships of the phenological stages, we propose to monitor three vegetative stages ('initial growth', 'leaves unfolding' and 'senescence') and two reproductive stages ('flowers open' and 'ripe fruits') to fully capture herbaceous species phenology.

**KEYWORDS**
first flowering day, flowering phenology, fruiting phenology, functional traits, growing season length, leaf out, senescence, vegetative phenology

## 1 | INTRODUCTION

Studies of phenology analyse the timing of biological events. In plant species, phenological events include leaf emergence, flowering, fruiting and leaf senescence. Phenology is of particular importance, as it is sensitive to climate change (Parmesan & Yohe, 2003; Walther et al., 2002). While the flowering times of herbaceous species have been well studied, there is relatively little data on the leaf and fruiting phenology of herbaceous species. It is estimated that over 50% of the worlds' species (FitzJohn et al., 2014) and c. 85% of the species of temperate ecosystems are non-woody (Ellenberg, 1996). Despite the huge proportion of herbaceous species, phenological research, particularly for leaf out and leaf senescence, has traditionally focussed on trees and shrubs or crops (Chmielewski & Rötzer, 2001; Panchen et al., 2014, 2015; Vitasse et al., 2011). Even for woody plants, most studies monitor leaf out and first flowering (König et al., 2018; Lechowicz, 1984; Panchen et al., 2014) rather than the later stages of phenology, such as the end of flowering, fruiting and leaf senescence (Bucher & Römermann, 2020, 2021; Gallinat et al., 2015; Panchen et al., 2015). Studies monitoring the entire life cycle of herbaceous plants are rare and limited to a few species (Cornelius et al., 2011; Ettinger et al., 2020; Kim et al., 2012).

Changes in plant phenology due to climate change are occurring all over the world and are already affecting biodiversity patterns and trophic interactions as well as ecosystem functions and services (Heberling et al., 2019; Inouye, 2008; Miller-Rushing et al., 2008; Rich et al., 2008). Many plant species respond to warming temperatures by altering the timing of phenological events (König et al., 2018; Menzel & Fabian, 1999; Root et al., 2003). Many studies report shifts towards earlier spring phenology and later autumn phenology, resulting in an overall increase in growing season length (e.g. Menzel & Fabian, 1999). Phenological shifts may also be caused by changes in precipitation regime (Crimmins et al., 2010; Fu et al., 2014; Huang, Koubek, et al., 2018), soil nutrients (Huang, Li, et al., 2018) and biotic interactions (Wolf et al., 2017). The magnitude and direction of phenological changes vary across species and depend on the season (Bucher et al., 2018; König et al., 2018; Parmesan, 2007), which may lead to changes in competitive hierarchies of ecosystems and to mismatches in biotic interactions (Heberling et al., 2019; Miller-Rushing et al., 2010).

The negative effects of mismatches on ecosystem processes are manifold: It has been pointed out, for example, by Heberling et al. (2019), Kudo et al. (2008), Lapointe (2001) and Lopez et al. (2008), that an earlier budburst of deciduous forest trees can lower the yearly photosynthetic rate of understorey herbs, especially in spring ephemerals. Plant–animal interactions such as plant–pollinator relations (Burkle et al., 2013; Kharouba et al., 2018; Kharouba & Vellend, 2015; Kudo & Cooper, 2019) or seed dispersal via animals (Gallinat et al., 2015) may be affected by species-specific responses.

There is widespread evidence that evolutionary history can be used to predict patterns in phenology, as closely related taxa tend to exhibit similar timing in their life-history events (Davies et al., 2013; Panchen et al., 2014, 2015). However, there is also evidence that phenological sensitivity to climate is not phylogenetically conserved and that closely related species do not show comparable flowering periods. However, the response to environmental changes is species-specific (CaraDonna & Inouye, 2015; Davis et al., 2010; Wolkovich et al., 2013). Recent studies suggest that plant functional traits determine phenological sensitivity and have the potential to improve predictions of phenological responses to environmental variation. For example, higher growth rates and leaf nitrogen content are associated with an earlier start of flowering in herbaceous species on local (Bucher et al., 2018) and global scales (König et al., 2018).

### 1.1 | Opportunities and challenges of monitoring phenology in botanical gardens

Botanical gardens offer a unique possibility to perform coordinated research on the effects of climate change (Primack & Miller-Rushing, 2009). Globally, there are 1,775 botanical gardens and arboreta in 148 countries on every continent except the Antarctic spanning wide climate conditions (BGCI, 2019). They encompass huge numbers of species in their collections that are phylogenetically, biogeographically and ecologically diverse. There is a large amount of overlap between gardens in species and in some cases gardens share clones (Chmielewski & Rötzer, 2001), an advantage when trying to separate the effects of genetic variation from environmental response. Because of their diverse collections, botanical gardens represent ideal locations to carry out evolutionary, ecological, biogeographical and morphological studies (Primack & Miller-Rushing, 2009).

However, there are also limitations associated with monitoring phenology in botanical gardens. It is often difficult to capture the effects of precipitation or soil nutrient conditions on phenology due to irrigation and fertilisation in the gardens. Other maintenance issues relate to fruit harvesting, pruning or weeding. Nonetheless, studies on phenology of trees in botanical gardens have successfully identified patterns in leaf out, senescence, fruiting and phylogenetic patterns of phenology for large numbers of woody taxa (Gallinat et al., 2018; Panchen et al., 2014, 2015). Botanical gardens have also been used to measure the timing and the speed of the growth of above-ground plant organs ('growth phenology') of large sets of herbaceous species to make the results independent of local differences in climatic drivers (Huang, Koubek, et al., 2018). As illustrated in Figure 1, phenological monitoring in botanical gardens offers the possibility to include many species from many habitats and different vegetation zones, reflecting the advantages of meta-analyses (see e.g. König et al., 2018; Root et al., 2003). At the same time, plant species grow under comparable (optimal) conditions in a relatively small area, allowing measurements of trait compositions and site conditions with a manageable effort, reflecting the advantages of local-scale field studies (see e.g. Bucher et al., 2018). In addition, studying plants in botanical gardens simplifies the monitoring of early season stages of vegetative phenology such as 'initial growth' which are hard to identify in a natural environment.

Here, we present the rationale of the PhenObs initiative—botanical gardens as a global phenological observation network—and lay out the development of a straightforward protocol that is appropriate for monitoring phenology across a vast diversity of growth forms of herbaceous species from various habitats and geographical regions.

### 1.2 | Monitoring phenology—Why do we need PhenObs?

Monitoring phenology has a long history. It was as early as the 9th century that recording the cherry blossom in Japan began (Aono & Kazui, 2008). In the 19th century, various European meteorological institutes started coordinated monitoring of plant phenology with a focus on woody species, often relying on the help of citizen scientists to gather data in the field (Austria: the Zentralanstalt für Meteorologie und Geodynamik (ZAMG) since 1851; UK: Royal Meteorological Society since 1875; Germany: Deutscher Wetterdienst since 1881). Today, we find a range of various initiatives and networks monitoring phenology, for example, IPG (International Phenological Gardens, 2019), the phenology program of the German Meteorological Service (Deutscher Wetterdienst, 2015), NEON (National Ecological Observatory Network; Elmendorf et al., 2016) and the USA-NPN (USA-NPN National Phenology Network Coordinating Office, 2013), partly monitoring also herbaceous species (e.g. USA-NPN). These initiatives and networks monitor the phenology mostly in wild populations supplemented by plants growing in private gardens (except for the IPG monitoring clones in gardens worldwide); some run also smaller projects in botanical gardens (USA-NPN). These networks collect an impressive amount of very valuable data that have also been included in various types of meta-analyses (e.g. König et al., 2018; Menzel & Fabian, 1999; Root et al., 2003).

Accordingly, we are establishing a new phenological network using botanical gardens (PhenObs) to monitor phenology on a large

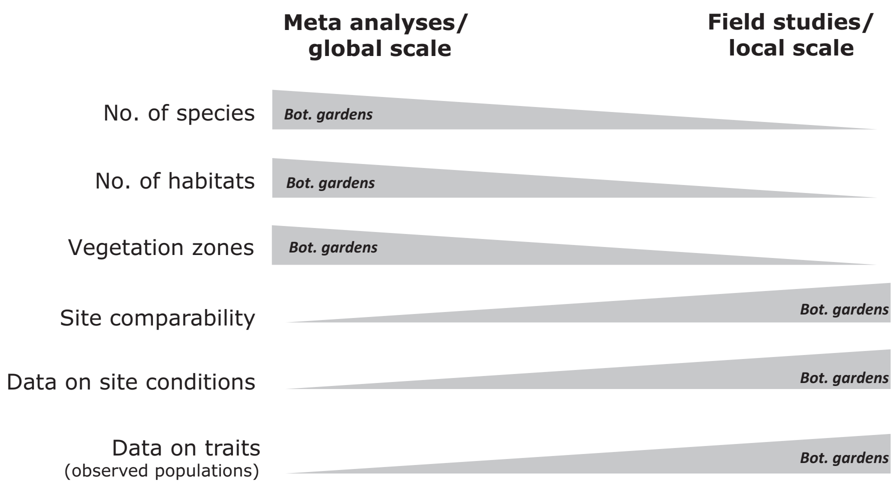


**FIGURE 1** Schematic diagram illustrating the advantages and constraints of global-scale and local-scale approaches to study plant phenology. The grey triangles indicate the amount of data (species, habitats, vegetation zones, site conditions, traits) and the degree of site comparability typically covered in global- versus local-scale approaches. Operating across botanical gardens enables the PhenObs network to profit from the advantages of both approaches, that is, to study large numbers of diverse taxa from diverse habitats and vegetation zones while incorporating site-specific information and traits measured on focal individuals/populations

set of herbaceous species from diverse families, habitats and geographical distributions in a standardised way. PhenObs (www.idiv.de/en/phenobs) is an open network of research groups in botanical gardens and includes several sites across the Northern hemisphere (Asia, North America, Europe). PhenObs researchers aim to improve the understanding of both vegetative (e.g. leaf out and senescence) and reproductive (e.g. flowering and fruiting) phenology focussing on herbaceous species, via standardised and coordinated phenological monitoring. This is in effect a large-scale 'common garden experiment' with replicates of species, genera and families grown around the world. The PhenObs network strives to answer the key questions:

1. What is the variation in plant phenology in herbaceous species across the growing season and in response to global change?
2. How can plant phenology be predicted from species' trait composition, provenance, position and extent of the distribution range and species' phylogeny?
3. What are the implications of this variation with respect to species performance and community assembly, biotic interactions (e.g. plant–pollinator interactions) as well as ecosystem processes and services, such as carbon sequestration and biomass production under changing land use and climate?

The network started monitoring in 2017 with a 2-year pilot phase conducted by four German botanical gardens (Berlin, Halle, Jena and Leipzig) to define a standardised and straightforward phenological protocol that can be applied to a large diversity of herbaceous species. One of our key initial questions was whether monitoring detailed substages of each phenological event was worth the effort and gave additional insights. We noted that a recent study by Ellwood et al. (2019) examining flowering, fruiting and leaf out of red maples in response to temperature across eastern North America using herbarium specimens showed that just a few phenological stages were sufficient to fully capture flowering, fruiting and leaf phenology. Similarly, Gallinat et al. (2018) showed for a range of woody plants at botanical gardens that the various stages of fruiting were highly correlated and that just recording first fruiting dates was sufficient for most purposes.

Here, we lay out the development of a standardised and straightforward protocol for observing vegetative and reproductive phenology across an extensive diversity of growth forms of herbaceous species from various habitats and geographical regions in botanical gardens. We conclude that a reduced number of clearly defined stages of phenology are sufficient to fully capture herbaceous species phenology based on the analyses of the relationship between 14 vegetative and reproductive stages monitored on more than 120 species in two subsequent years.

## 2 | MATERIALS AND METHODS

### 2.1 | Study sites and environmental parameters

The study was conducted at the botanical gardens of Berlin, Halle, Jena and Leipzig in 2017 and 2018, being located in Eastern and Central Germany in a subcontinental climate (see Table 1). Note that 2018 was about two degrees warmer than 2017, and both years were warmer the average of 1981–2010.

Further gardens covering a large geographical gradient and climate extent across the Northern hemisphere have joined the network since 2019 (see www.idiv.de/en/phenobs for a most recent overview on network members).

### 2.2 | Species selection and plot design

We selected up to 170 herbaceous species for our study reflecting a high diversity in growth forms, ecological niches, phylogeny and being widely available in the four gardens. The species list is in Supporting Information 1. Not all species were present in every garden, but in 2018 51% of the species were monitored in at least three gardens and 24% of the species in all four gardens. To cover a larger variability of growth forms occurring in the herbaceous layer, we added some dwarf–shrub and subshrub species to our list (e.g. *Solanum dulcamara*, *Gaultheria shallon*, *Vaccinium myrtillus*).

We defined a population as a group of a particular species regardless of how many individuals were present. Some of these populations originated from seeds collected in the field, whereas others were clones from one or a couple of individuals. In the case of the few chamaephytes (dwarf–shrub and subshrubs), data sometimes referred to a single individual.

Observations were made over the entire population for each species. As the area of each species cover varied from one individual (in

**TABLE 1** Botanical gardens in which plant phenology was monitored in the pilot study years with their location, their climate conditions and the numbers of species observed in 2017 and 2018. Temperature data come from the DWD Climate Data Center (CDC) (2020)

| Botanical garden | Latitude, longitude | Altitude [m a.s.l.] | Mean annual/ March/May temp. [°C], 1981–2010 | Mean annual/ March/May temp. [°C], 2017 | Mean annual/ March/May temp. [°C], 2018 | Number of species analysed 2017/2018 |
|---|---|---|---|---|---|---|
| Berlin | 52°27′N, 13°18′E | 50–70 | 9.5/4.7/14.3 | 10.1/7.3/14.9 | 11.1/1.7/17.6 | 125/170 |
| Halle | 51°29′N, 11°57′E | 85–105 | 9.6/5.0/13.9 | 10.5/8.0/14.6 | 11.3/2.5/17.0 | 66/94 |
| Jena | 50°55′N, 11°35′E | 150–170 | 9.9/5.4/14.3 | 11.0/8.5/15.8 | 11.5/3.3/17.1 | 78/130 |
| Leipzig | 51°19′N, 12°23′E | 120 | 9.4/4.7/13.8 | 10.5/8.0/15.2 | 11.4/2.8/17.0 | 0/94 |

the case of *G. schallon*) up to several m$^2$ (e.g. *Aegopodium podagraria*), we tried to do the observations always on the same localised (though not marked) place, generally monitoring an approximately 1 m$^2$ plot per species in each garden. This localised monitoring was done to ensure uniform environmental conditions at this place. For some species, plants were growing in mixtures with other species. Within the PhenObs protocol, we recorded all site characteristics for each species to account for differences that may be associated to differences in monitoring area and plant densities (analyses not included here). However, based on our extensive experience, such site differences are far less important than the large differences among species.

### 2.3 | Monitoring protocols

For each species, 14 vegetative and reproductive phenological stages covering the seasonal life cycle were monitored (Table 4). For the flowering stages, the intervals were particularly fine scaled, including the onset of buds and the percentage of open flowers (first flower, <10%, <50%, last 10%, no flowers open), to reflect the development of flowers in detail (see Supporting Information 2a for an example data entry sheet).

Table 4 gives an overview on the definition of the stages we monitored. The stages were selected based on expert opinion regarding feasibility and application to herbaceous species. These are similar to the stages used in the BBCH system (Biologische Bundesanstalt für Land- und Forstwirtschaft, 2001; Meier et al., 2009) as applied by the German Meteorological Service (DWD, Deutscher Wetterdienst, 2015), by Denny et al. (2014) as well as the stages used by the USA-NPN (USA National Phenology Network Coordinating Office, 2013). Many of these stages are also observed through the IPG network (International Phenological Gardens, 2019) and are similar to the stages adapted by NEON (U.S. National Earth Observation Network; Elmendorf et al., 2016).

We selected vegetative and reproductive stages and modified the definition of stages to deal with the diversity of growth forms in herbaceous plants while keeping them as straightforward as possible to allow volunteers to participate in the project after some initial training. In some groups of species, it may be difficult to observe some of the proposed stages (e.g. first leaf visible in its typical form in the case of sedges). For these cases, the monitoring sheets provide the possibility to enter 'unsure' so that this data are not—or only after plausibility check—included in further analyses (for more details, see also Supporting Information 4: PhenObs protocol). In the pilot phase, for all stages, the date of the onset of the stages were recorded as soon as observed on at least one plant—or on three twigs of one individual in case of chamaephytes.

We monitored phenology weekly on a population level as recommended by Cornelius et al. (2011), Miller-Rushing et al. (2008) and Panchen et al. (2014).

The data are publicly available via the iDiv data repository: https://doi.org/10.25829/idiv.1877-4-3160 (Nordt et al., 2020).

### 2.4 | Data analyses

To identify which stages are necessary to fully capture plant phenology of herbaceous species, we tested the relationship of the dates of different vegetative and reproductive stages. All species monitored in at least one of the four gardens were included in the analysis (see Supporting Information 1: species list).

For the analyses of vegetative stages, we tested whether the day of the year of the 'first leaf' (the day, when the form of the newly formed leaf was recognisable) can be used to predict the other vegetative stages ('initial growth', 'leaf unfolded', 'onset of senescence' and 'peak senescence'). We performed an analysis of covariance (ANCOVA) with the day of the year (DOY) of the 'first leaf' as predictor variable and garden and year included as covariates alone and in interaction with 'first leaf'. Full models were simplified via backwards selection until the minimum adequate model was found as described in Crawley (2012). As continuous response variable, we included each of the other vegetative stages in separate models (see Table 4). In addition, we tested whether the stage 'onset of senescence' can be used as a proxy also for 'peak senescence' using the same model as described above. We computed the fraction of variation attributable to each predictor variable in the models using the calcVarPart function (Hoffman & Schadt, 2016).

As day of 'first flower' is the most frequently used reproductive stage in phenology research, we tested whether the day of the year of the 'first flower' can be used to predict other reproductive stages. As for the vegetative stages, we conducted an ANCOVA with 'first flower' as predictor and each of the other reproductive stages (see Table 4) as response variable in separate models. Also here, we included garden and year as covariates alone and in interaction with 'first flower'. In the 'flower buds swollen' models, Leipzig garden was excluded due to missing data. For the same reasons as outlined above for the senescence stages, we additionally tested whether 'first ripe fruit' captures 'peak fruiting' using the same modelling approach.

All statistical analyses were conducted in R (R Core Team, 2019) with the R package VARIANCEPARTITION (Hoffman & Schadt, 2016). All plots were produced using the R package GGPLOT2 (Wickham, 2009).

## 3 | RESULTS

### 3.1 | Relationship between 'first leaf' and the other vegetative stages

The results of the models for the vegetative phenological stages showed that the phenological stage 'first leaf' was highly correlated with 'initial growth' and 'leaf unfolded' (Figure 2a,b, for statistics, see Table 2). The variable 'garden' had a significant effect on the intercept in the leaf unfolded model, and year had a significant effect on the slope of both models. The association between 'first leaf' and the two senescence stages was much less strong though still significant (Figure 2c,d,

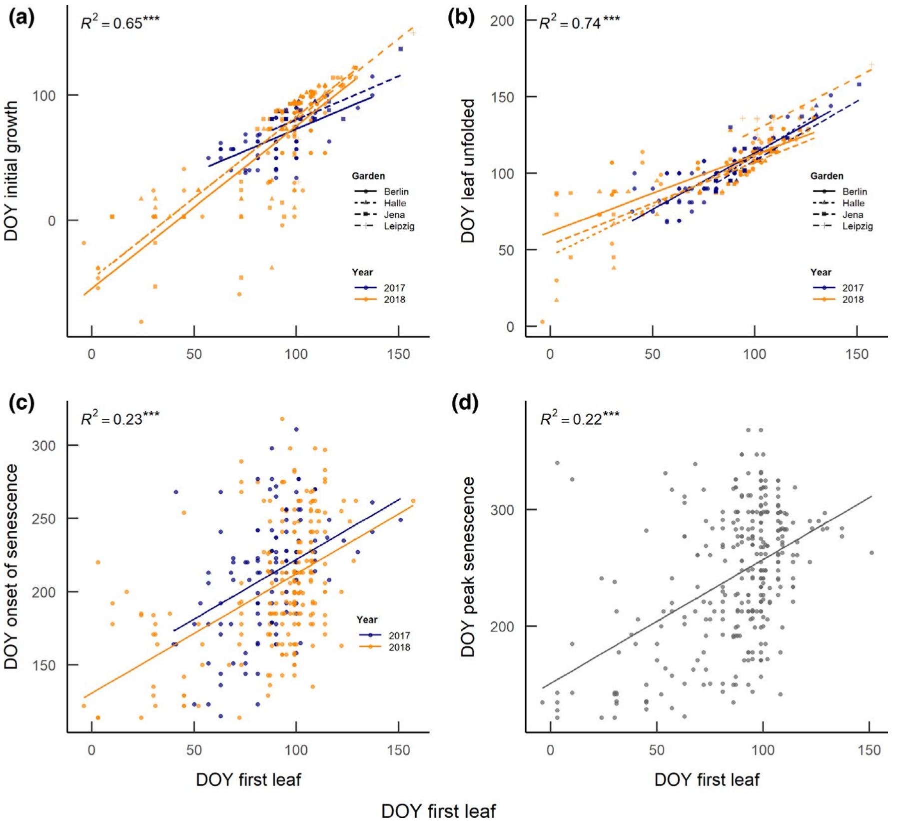


**FIGURE 2** The relationship between the day of the year (DOY) of first leaf with the vegetative stages initial growth (a), leaf unfolded (b), onset of senescence (c) and peak senescence (d). Each dot represents the observation of one species in one garden and one year. Lines result from the ANCOVAs where each phenological stage was a continuous response variable predicted by first leaf as a continuous predictor and garden and year as categorical covariates alone and in interaction with DOY first leaf. In models that showed significant effects of covariates, different colours indicate years, different symbols indicate gardens. For statistical test values, see Table 2

**TABLE 2** Results of the linear regression models using the day of the year of the phenological stage first leaf to explain the day of the year of initial growth, first leaf unfolded, onset of and peak senescence. Garden and year were included alone and in interaction with first leaf as covariates in the models. Given are the outputs of the model ($R^2$, $F$-statistics). Significant effects are indicated with $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$, n.s. not significant. '—' indicates terms that were not included in the minimal adequate models. Percentages give the fraction of variation attributed to each variable included in final model based on variance partitioning methods

| | Initial growth | Leaf unfolded | Onset of senescence | Peak senescence |
|---|---|---|---|---|
| | $R^2 = 0.65$*** | $R^2 = 0.74$*** | $R^2 = 0.23$*** | $R^2 = 0.22$*** |
| | $F_{6,318} = 99.9$*** | $F_{9,433} = 139.7$*** | $F_{2,354} = 51.6$*** | $F_{1,309} = 86.8$*** |
| First leaf | *** (65%) | *** (73%) | *** (22%) | *** (22%) |
| Garden | n.s. (1.6%) | *** (13%) | — | — |
| Year | n.s. (0.1%) | n.s. (0.2%) | * (2%) | — |
| First leaf × garden | — | n.s. (1%) | — | — |
| First leaf × year | *** (5%) | *** (5%) | — | — |

Table 2), with a significant effect of year on the intercept in the onset of senescence model. The two senescence stages, onset of and peak senescence were highly correlated with one another ($R^2 = 0.64$, $F_{5,394} = 142.7$, $p < 0.001$), with a significant effect of garden and year on the intercept (for both $p < 0.001$; see Supporting Information 3).

## 3.2 | Relationship between 'first flower' and other reproductive stages

As presented in Table 3 and Figure 3, there were strong relationships between 'first flower' and the other reproductive phenological stages selected; there was no consistent effect of garden and year in the models. The relationship between 'first flower buds' and 'first flower' was strong (Figure 3a) with no significant effect of garden and year. The relationship between 'flower buds swollen' and 'first flower' was even stronger, but both, garden and year had an effect on the slope of the regression lines (Figure 3b). 'Flowers 10% open' and 'first flower' were strongly associated to one another though there was a significant year effect on model slope (Figure 3c). The association between 'peak flowering' and 'first flower' was strong and differed between gardens and years (Figure 3d). The relationship between the last 10% of flowers being open ('last 10% flowering') and 'first flower' was significant, again with effects of garden and

**TABLE 3** Results of the linear regression models using the phenological stage 'first flower' (FFD) to explain six other flowering stages and two fruit stages. Garden and year were included alone and in interaction with FFD as covariates in the models. Given are the outputs of the model ($R^2$, $F$-statistics). Significant effects are indicated with $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$, n.s. not significant. '–' indicates terms that were not included in the final models. Percentages give the fraction of variation attributed to each variable included in final model based on variance partitioning methods

| | First flower buds | Flower buds swollen | Flowers 10% open | Peak flowering | Last 10% flowering | End flowering | First ripe fruit | Peak fruiting |
|---|---|---|---|---|---|---|---|---|
| $R^2$ | 0.85*** | 0.97*** | 0.99*** | 0.96*** | 0.81*** | 0.66*** | 0.59*** | 0.56*** |
| | $F_{1,412} = 2,391^{***}$ | $F_{1,434} = 16,760^{***}$ | $F_{3,501} = 15,000^{***}$ | $F_{9,502} = 1,359^{***}$ | $F_{2,470} = 1,013^{***}$ | $F_{4,474} = 225.9^{***}$ | $F_{2,351} = 253.9^{***}$ | $F_{2,420} = 269.2^{***}$ |
| FFD | *** | *** | *** | *** | *** | *** | *** | *** |
| | (85%) | (87%) | (98.9%) | (96%) | (81.1%) | (65.3%) | (56.4%) | (55.1%) |
| Garden | – | – | – | ** | – | ** | – | – |
| | | | | (2.9%) | | (2.8%) | | |
| Year | – | – | n.s. | n.s. | * | – | ** | * |
| | | | (0.41%) | (0.01%) | (1.2%) | | (2.1%) | (1.2%) |
| FFD × garden | – | – | – | * | – | – | – | – |
| | | | | (1.6%) | | | | |
| FFD × year | – | – | * | ** | – | – | – | – |
| | | | (0.87%) | (1.5%) | | | | |

year on the slopes. 'End of flowering' was significantly associated with 'first flower', with a significant difference of the intercepts of the gardens. Both fruiting phenological stages were strongly and significantly associated with 'first flowering day', though these relationships were weaker than for the flowering stages. In both cases, year had an effect on the intercept (Figure 3g,h). Concerning the association between the two fruiting stages the models suggest that 'first ripe fruit' and 'peak fruiting' were significantly correlated ($R^2 = 0.87$, $F_{9,431} = 597.2$, $p < 0.001$), with effects of garden ($p < 0.05$) and year ($p < 0.05$) on the intercept (see Supporting Information 2).

## 4 | DISCUSSION

Our study shows that although the phenology of species differed between years and gardens, many phenological stages are tightly related. The effects of garden and year on the relationships between the stages were very small, typically not explaining more than 5% of model variation. Thus, based on our analysis, we show that a reduced set of phenological stages can be used to sufficiently characterise plant phenology to assess changes with regard to climate and associated shifts in ecosystem functions. As such, we suggest a new standardised monitoring protocol focussing on a few selected phenological stages that allow monitoring a large set of species with reasonable efforts (Supporting Information 4).

### 4.1 | Developing standardised protocols for herbaceous species

Table 4 gives an overview of the stages we observed in the pilot study alongside the new, reduced set of stages, which we propose for monitoring the phenology of a large set of herbaceous species. We recommend to monitor two vegetative stages: (1) 'Initial growth' indicates the start of the growing season. Even though it is highly correlated with 'first leaf', slopes and intercepts differed between gardens and years, suggesting to keep this stage. (2) 'Young leaves unfolding' indicates the time when species typically begin photosynthesis, at least in temperate regions. Here, we renamed the term 'first leaf', which was used in the pilot study to account for the possibility to include a second leaf flush during the growing season. By monitoring these two stages early in the season, that is, when new shoots appear or young leaves unfold, we will be able to provide novel data for phenology research, as these stages are inherently difficult to monitor in the wild (e.g. Cornelius et al., 2011). Especially in dense vegetation types in the wild, it is difficult and sometimes nearly impossible to distinguish plant species in very early stages in spring (unless detailed information on species composition is available from previous year), underlining also the advantage of monitoring herbaceous plant species in botanical gardens. With the PhenObs initiative, we will be able to provide data on these early-season vegetative stages which indicate the start of the growing season and which allow calculating growing season length. Eventually, we can test

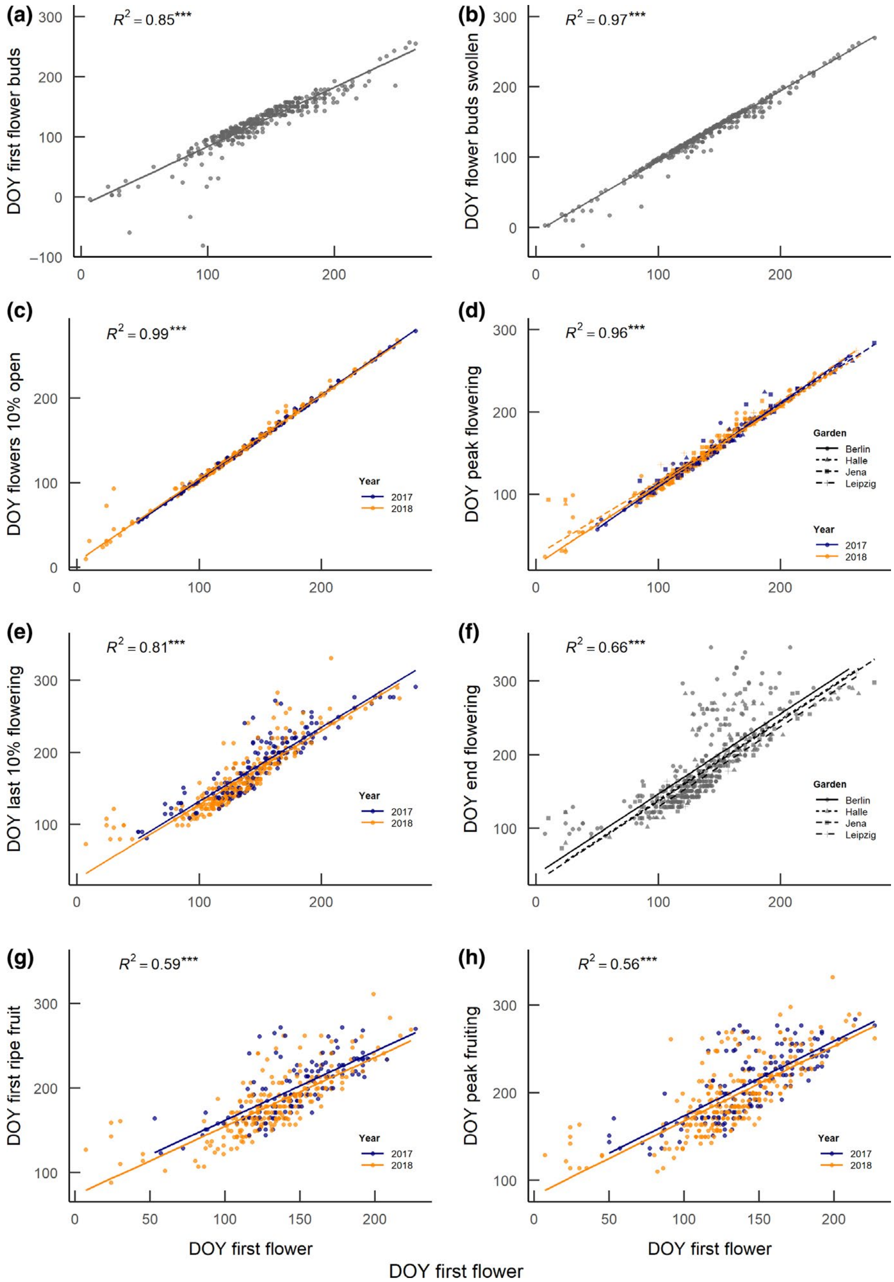


**FIGURE 3** The relationship between first day of year (DOY) of the flowering stages flower buds (a), flower buds swollen (b), flowers 10% open (c), peak flowering (d) last 10% flowering (e), end flowering (f), first ripe fruit (g), peak fruiting (h) and DOY of first flower resulting from the models where each phenological stage was a continuous response variable predicted by first flower as a continuous predictor and garden and year as categorical covariates alone and in interaction. In models that showed significant effects of covariates, different colours indicate years, different symbols indicate gardens. For statistical test values, see Table 3

whether data on these early-season phenological stages can be predicted from (easy-to-measure) data on plant traits, either measured on the same populations (as planned in e.g. PhenObs, see Protocol in Supporting Information 4), or, alternatively, from trait databases such as TRY (Kattge et al., 2020).

We also suggest monitoring (3.1) 'leaf senescence' as an additional vegetative stage as the observed correlation between 'first leaf' and 'leaf senescence' was rather low (Table 4). The highly significant relationship between 'onset of senescence' and 'peak senescence' allows extrapolating these dates from one another. However, to capture effects of, for example, summer drought within the growing season we propose to estimate (3.2) senescence intensity throughout the year. We further suggest monitoring when at least 50% senescence is reached at the end of the growing period, as this 'peak senescence' was also proposed in other studies and networks to define the end of the growing season (e.g. Budburst, 2019; Deutscher Wetterdienst, 2015; International Phenological Gardens of Europe, 2019; Panchen et al., 2015). From the vegetative stages, which we propose to monitor, we are able to specifically capture the length of the growing season, which is of considerable value in global change research. Changes of growing season length have implications for the global carbon cycle and ecosystem functions like productivity, as well as species performance and community composition (Churkina et al., 2005; Kramer, 1995; Richardson et al., 2010; White et al., 1999). Furthermore, changes in the growing season length may affect biotic interactions such as herbivory: when photosynthetic tissues are produced and leaves are greening up they also become most susceptible to herbivory. Climate-induced changes of the appearance of leaf tissue have thus also implications for interactions

**TABLE 4** Development of a simplified protocol to monitor phenology that is applicable to a large set of herbaceous species. Shown is a comparison of the 14 phenological stages that have been monitored during the pilot phase 2017/2018 and the proposed reduction to five stages for the PhenObs network and beyond. A full description of the PhenObs protocol can be found in Supporting Information 4; its most recent version (including FAQ section) and via www.idiv.de/en/phenobs

| | Full version (pilot study) | Simplified version (PhenObs) |
|---|---|---|
| Vegetative phenology | **1. Initial growth**<br>First appearance of the shoot above-ground*<br>• Noted only at first sight.<br>• Considering any growth irrespective of its origin (shoot, leaf, leaf-sheath, flower). | **1. Initial growth**<br>First appearance of the shoot aboveground*<br>• Noted as long as new shoots appear.<br>• Considering only vegetative growth. |
| | **2a. First leaf**<br>The first leaf is fully visible in its typical form (can still be partly folded). It should usually be green.<br>• Noted only at first sight.<br>• Second leaf flushes are noted as remark. | **2. Young leaves unfolding**<br>The first leaf is fully visible in its typical form (can still be partly folded). It should usually be green.<br>• Noted at every sight, including also second leaf flushes during the growing season. |
| | **2b. First leaf unfolded**<br>The first leaf is unfolded and has at least 75% of its size.<br>• Noted only at first sight. | *<highly correlated with young leaves unfolding>* |
| | **3a. Onset of senescence**<br>Leaves are changing colour, dry out or fall off.<br>• Noted only once.<br>• Dry leaves as result of any pest infestation or summer drought are not evaluated as senescent. | **3.1 Senescence**<br>Leaves are changing colour, dry out or fall off.<br>• Noted continuously when senescent leaves are sighted.<br>• Dry leaves as a result of summer drought or pest infestation can be included.<br>**3.2 Senescence intensity****<br>• The additional estimation of percentage senescence ('intensity') allows calculating all possible thresholds, including 'peak senescence'. |
| | **3b. Peak Senescence (50%)**<br>• At least 50% of the leaves have fallen off, dried or coloured.<br>• Noted only once. | *<can be extracted from the intensity column>* |
| Reproductive phenology | **4a. First flower buds**<br>The first flower bud is visible.†<br>• Noted only once. | *<highly correlated with first flower>* |
| | **4b. Flower buds swollen**<br>Buds are swollen; the colour of the flowers can be recognised.<br>• Noted only once. | *<highly correlated with first flower>* |
| | **4c. First flower**<br>First flower is fully open<br>• Noted only once. | **4.1 Flowers/inflorescences open**<br>Start monitoring when first flower is fully open.<br>• Indicating 'yes' for every week flowering is observed.<br>**4.2 Flower intensity‡**<br>• The additional estimation of percentage flower intensity allows calculating all possible thresholds, including 'peak flowering', 'end of flowering', etc. |
| | **4d. Flowers 10% open**<br>At least 10% of the flowers/inflorescences are open.<br>• Noted only once. | *<can be extracted from the intensity column>* |
| | **4e. Peak flowering (50%)**<br>At least 50% of the flowers are open. The maximum number of flowers opening at the same time in each population is defined as the 100% standard.<br>• Noted only once when 50% is reached. | *<can be extracted from the intensity column>* |
| | **4f. Last 10% flowering**<br>Less than 10% of the flowers are open. | *<can be extracted from the intensity column>* |

(Continues)

**TABLE 4** (Continued)

| | Full version (pilot study) | Simplified version (PhenObs) |
|---|---|---|
| | **4g. End of flowering (0%)**<br>No more flowers are open (stamen and stigma are withered). | *<can be extracted from the intensity column>* |
| | **5a. First ripe fruit**<br>Onset date of first ripe fruit.<br>• Noted only once when first fruit is ripe. | **5. Ripe fruit**<br>Start monitoring when first fruit is ripe, indicating 'yes' for every week a ripe fruit is observed. |
| | **5b. Peak fruiting**<br>The date when at least 50% of the fruits are ripe, overripe or fallen off. | *<highly correlated with first fruit>* |

*Special cases: (a) overwintering buds: the first tip of photosynthetic tissue emerging from the bud in spring. (b) chamaephytes: bud swelling; (c) wintergreen hemicryptophytes: initial growth from ground, not new leaves on the old stems (for more details, see Supporting Information 4).

**Senescence intensity is an optional category in PhenObs. In any case, at the end of the vegetation period, monitoring stops when 50% is reached.

†Special cases: In case of very dense inflorescences like the heads of Asteraceae, the visibility of the inflorescence bud should be considered (for more details, see Supporting Information 4).

‡Flower intensity is an optional category in the PhenObs project, though we recommend to at least capture 50% flower intensity.

with herbivores (Aizen & Patterson, 1995; Mayor et al., 2017; Post & Forchhammer, 2008; Renner & Zohner, 2018).

Concerning the reproductive stages, we recommend to observe at least 'first flower', as this stage was closely related to the other flowering stages. Our experience from the pilot study enabled us to change the type of data recording from event-based (only onset dates) to status-based monitoring, as described in Denny et al. (2014). We thus recommend to weekly record a stage as long as it is visible. In the case of 'open flowers' (which replaces the term 'first flower', see Table 4), these data allow extracting information on second or third flushes and flowering duration (compare also e.g. Bucher & Römermann, 2020).

Throughout the course of the year and with the progression of flowering, the association between first flowering day and the other reproductive stages became weaker. We therefore suggest capturing changes in flowering patterns with the additional monitoring of (4.2) 'flower intensity' to record the percentage of open flowers. From these data, peak flowering can be extracted. Flowering duration and peak flowering represent important phenological phases and stages influencing ecosystem performance, for example, the availability of nectar and pollen for pollinators and therefore the reproduction success of the respective plant species (Burkle et al., 2013; Forrest, 2014; Kudo & Cooper, 2019; Kudo et al., 2008). The observation that the correlations between flowering stages changed in the course of the year were also confirmed by other studies: CaraDonna et al. (2014) showed, on 60 species in a subalpine plant community, that first, peak and end flowering shifted independently of one another. Bucher and Römermann (2020) confirmed that changes in first and last flowering day as well as second flowering flushes followed different patterns along elevational gradients according to different strategies on a set of 29 herbaceous species.

We further suggest to monitor (5) 'ripe fruit', as 'first flower' was only loosely related to the fruiting stages. As 'first fruit' was highly correlated with 'peak fruiting' (Supporting Information 2), the observation of this latter stage would not provide additional information. Accordingly, we propose similar to the flowering stages to record if a plant displays ripe fruits (or not) as this is more straightforward to monitor. The availability of ripe fruits may, however, be observed through the entire wintertime, as this can be of special interest when aiming to study the influence of fruit display on bird migration and feeding (Gallinat et al., 2018; Knudsen et al., 2011) and to deduce the length of the dispersal period and its possible influence on reproductive success of the species. However, maintenance issues in botanical gardens and exhaustive seed collections for the Index Seminum are likely to result in missing data especially for 'peak fruiting', but the correlation between 'first ripe fruit' and 'peak fruiting' suggest that the latter can be extrapolated from 'first ripe fruit'.

A detailed and illustrated description of the different stages is provided in the PhenObs protocol (Supporting Information 4). In this protocol, we specifically refer to the diverse growth forms which can be problematic in evaluating the stages and making them comparable between species, for example, the 'young leaves unfolding' in monocotyledons or 'initial growth' in rosette forming species. Additional examples of phenological stages and special cases are given, described and illustrated there.

## 4.2 | Advances to monitoring phenological stages

The experience of the detailed monitoring of different stages capturing the vegetative and reproductive phenology led us to switch from monitoring only the onset of each phenological stage to recording its duration and, partly, intensity similar as described in Denny et al. (2014) (also called status-based monitoring). With this approach, we propose to record all days during which a phenological event is visible and thus capture both the beginning and end of a phenological stage from which the duration of a phase and also peak in case of flowering and senescence can be derived. With this approach, it is possible to record multiple events of the same phenological phase within a year, for example, two distinct leaf flushes or flowering periods or peaks, that are inherently hard to capture though they show clearly species-specific patterns (Bucher & Römermann, 2020). These phases are important to inform predictions on the effect of changes in the abiotic and biotic environment on species diversity.

The standardised protocols (Supporting Information 4) can be applied across diverse herbaceous species. We provide guidance on the protocol as well as regular updates on the project homepage (www.idiv.de/en/phenobs). Designing a simple and straightforward protocol opens up application by volunteers to contribute to the project by gathering large datasets in any other gardens of the world and thus amplify the relevance of botanical gardens and open climate change research to a broader public. This protocol also opens the possibility with links to existing (or future) Citizen Science projects (such as e.g. BudBurst in the United States or Natuurkalender in the Netherlands, though they monitor in the wild).

## 5 | CONCLUSIONS

With this study, we have developed a standardised monitoring protocol as a basis of the PhenObs network to capture species-specific differences in the phenology of herbaceous plant species. We identified a minimum set of stages that accurately capture both the vegetative and reproductive phenology of herbaceous plants. These stages are relevant for global change studies and its effects on ecosystem productivity, ecosystem services, above-ground biotic interactions, species performance and community composition.

With the establishment of PhenObs, we aim to fill the gap of knowledge of herbaceous species phenology by analysing phenological stages as a function of their traits on a large set of herbaceous species in a controlled setting and characterise the environmental (micro-)site conditions to study the effect of changing climate on species performance. PhenObs continues monitoring and aims at extending the number of partner gardens using the standardised protocols presented here (Supporting Information 4) for monitoring phenology, combined with the measurement of functional traits. Included traits reflect key aspects of a species performance in different environments and were identified in previous studies as relating to phenology (Bucher et al., 2018; König et al., 2018), such as plant height, specific leaf area or leaf nitrogen content. Differences between gardens that have been shown in this pilot study may be related to differences in maintenance issues, garden-specific species sets and differences in the starting times of yearly observations of the four gardens that bear otherwise comparable climate conditions. The future expansion of the PhenObs network covering more diverse climate conditions will allow exploring ecological and climate factors associated with the variation in vegetative and reproductive phenology in herbaceous species. Once these data are collected, the PhenObs network will further identify the implications of variation in phenology with respect to species performance and assembly and biotic interactions (e.g. plant–pollinator interactions) as well as ecosystem processes and services under global change. Overall, we hope this standardised protocol will serve as useful tool to study phenology shifts in a changing climate and thereby unravel the effects on ecosystem functions and services. By developing the open PhenObs network, we also aim to instigate a backbone for climate change research that is also accessible to the general public and thereby help foster a greater understanding of climate-induced consequences on ecosystems and biodiversity.

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### AUTHORS' CONTRIBUTIONS

A.B., S.F.B., M.F., I.H., B.N., R.B.P., C.R., A.-D.S. and C.W. (in alphabetical order) conceived the ideas of the PhenObs project; S.F.B., M.F., I.H., B.N., C.P., C.R. and A.-D.S. designed the methodology of the protocols and, together with D.J. and M.S. critically revised and adapted previous versions of the protocol taking into account inconsistencies that were realised during the monitoring in the Botanical Gardens in the pilot phase of the PhenObs project; M.F. and B.N. led and C.P. supported the data collection in Leipzig, Berlin and Halle, respectively; in Jena, data collection was established and later supervised by C.R. and S.F.B. C.R., B.N. and M.S. analysed the data; B.N., C.R. and I.H. led the writing of the manuscript. All authors contributed critically to the drafts and gave final approval for publication.

### DATA AVAILABILITY STATEMENT

The data are publicly available via the iDiv Data Repository https://doi.org/10.25829/idiv.1877-4-3160 (Nordt et al., 2020).

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## SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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