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Substrate composition impacts long-term vegetation development on blue-green roofs: Insights from an experimental roof and greenhouse study

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ABSTRACT

Green roofs provide ecosystem services and can promote biodiversity in urban areas. Blue-green roofs have an additional water storage compartment under the substrate to reduce roof water runoff, thereby also reducing drought stress which is beneficial for green roof vegetation. In order to study which blue-green roof design supports the highest plant diversity, we assessed the effect of different substrates and seed mixtures on vegetation development in a short-term greenhouse experiment and long-term blue-green roof experiment. A ten-week full-factorial greenhouse experiment was performed for six substrate composition and four seed mixture treatments. On an experimental blue-green roof, we annually surveyed plants from 2013 to 2021 in nine different treatments (five replicates each), that varied in substrate composition, substrate depth and seed mixture that was initially applied. Two treatments resembled conventional non-green roofs (100% gravel) and conventional extensive *Sedum* green roofs. The results of the greenhouse experiment showed that seed mixture is more important than substrate composition in shaping the initial species richness and species composition. However, on the experimental roof the substrate composition was an important determinant of species richness and species composition long-term. Plant species richness on the experimental roof was lowest in the gravel treatment (resembling conventional non-green roofs), and highest in treatments where locally collected soil was used, likely due to additional species that appeared from the seed bank present in the transplanted soil. Soil was never completely covered with vegetation on unfertilized substrates that contained 20% or less dense and organic materials. Plant species richness on conventional *Sedum* roof substrate was higher on the experimental blue-green roof compared to an adjacent non-blue roof, highlighting that blue-green roofs can promote biodiversity more than conventional green roofs. For future construction of blue-green roofs in our region, we recommend the addition of 30% locally collected soil to a 6 cm deep lightweight substrate to maximize long-term plant cover and plant species richness.

1. Introduction

Green roofs benefit people and nature since they provide ecosystem services and enhance biodiversity in urban areas (Berardi et al., 2014; Madre et al., 2014; Williams et al., 2014; Oberndorfer et al., 2007). As a result, the application and research on green roofs has grown enormously over the last two decades (Blank et al., 2013). Ecosystem services provided by green roofs include reduction of the urban heat island effect, reduced energy consumption of buildings and reduced rainwater runoff (Mentens et al., 2006; Francis and Jensen, 2017). Although there is uncertainty whether green roof biodiversity can match ground-level biodiversity, there is little doubt that arthropod and plant diversity is greater on green roofs than on conventional roofs (Williams et al., 2014).

The design of a green roof influences the plant diversity it can support. Typically, the depth and composition of the substrate has a large influence on plant growth (Dvorak and Volder, 2010; Rowe, 2015; Kazemi and Mohorko, 2017). Deeper substrates generally promote plant diversity since they provide more structure for roots, contain more nutrients and retain more moisture. Shallow substrates are often dominated by drought-resistant succulents, whereas deeper substrates support more herbaceous plants (Durhman et al., 2007; Brown and Lundholm, 2015; Gabrych et al., 2016; Van der Kolk et al., 2020). Consequently, roofs with a thin substrate layer (extensive green roofs)

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generally only support a *Sedum* vegetation, whereas roofs with a deep substrate layer (intensive green roofs) can support forbs, bushes or even trees (Berndtsson et al., 2009; Berardi et al., 2014).

Extensive green roofs are typically constructed using a light-weight substrate such as volcanic stone, which does not break down over time and does not cause nutrients to runoff (Rowe et al., 2006; Rowe, 2015). However, light-weight substrates are nutrient-poor and their water holding capacity is limited (Nagase and Dunnett, 2011). Consequently, especially non-succulent plant species often require substrates with higher organic content (Rowe et al., 2006; Young et al., 2014). In order to promote growth of non-succulent plant species, organic material such as peat and compost can be mixed with light-weight material (Rowe et al., 2006). Another option is to apply local soil on green roofs, and this may result in very diverse roof vegetations since seeds present in the soil may boost plant species richness. For example, the 30,000 m^2 green roof of the Moos water filtration plant near Zürich (Switzerland) was constructed in 1914 using a deep layer of sand and local soil, and harbours currently 175 plant species (Rowe, 2015).

Drought is often limiting plant growth on green roofs and especially on extensive roofs with a shallow substrate layer (Dvorak and Volder, 2010; Olly et al., 2011; Bates et al., 2013; MacIvor et al., 2013). Water availability throughout the year can be increased by constructing an additional water storage compartment under the green roof substrate layer. Roofs with such a design ("*blue-green roofs*") may thus not only be beneficial because they reduce rainwater runoff more than conventional green roofs (Shafique et al., 2016), but also because they may support higher plant diversity by reducing drought stress.

Long-term vegetation studies on (blue-)green roofs are essential to understand which roof designs optimize plant diversity. There are, however, still relatively few studies that examine how substrate composition affects vegetation development on green roofs in a field setting and over longer time-periods (Bates et al., 2015; Vandegrift et al., 2019; Zhang et al., 2021), and, to the best of our knowledge, no study has done this for blue-green roofs.

We performed a short-term greenhouse experiment and a long-term blue-green roof experiment to study vegetation development on blue-green roofs. The ten-week full-factorial greenhouse experiment aimed to study the relative impact of substrate composition and seed mixture on initial plant species richness and plant biomass. The blue-green roof experiment, performed on the building of the Netherlands Institute of Ecology over a nine-year period, was designed to study how vegetation characteristics varied among nine treatments that differed in their substrate composition and initial seed mixtures that were applied. The nine treatments were selected so that they include a variety of systems that resemble conventional black and green roofs, and varied in the proportion of dense materials used in the substrate. Some treatments included substrates containing locally collected soil to boost plant diversity. The main goal of this study was to explore which blue-green roof designs promote plant cover and plant diversity, knowledge that can be applied in the design of new blue-green roofs to optimize their potential for biodiversity. Additionally, this study provided an excellent opportunity to compare plant species richness and composition between a blue-green roof and a conventional green roof, that is situated on the same building and was studied over the same time period (Van der Kolk et al., 2020).

2. Methods

2.1. Study site and setup

2.1.1. Greenhouse experiment

Due to the limited roof space it was not possible to study in-situ all combinations of substrates and initial seed mixture compositions. A greenhouse experiment was therefore performed to study the relative importance of substrate and seed mixture composition on short-term plant species richness and presence of different functional groups. We performed a full-factorial greenhouse experiment for six different substrate treatments and four seed mixture treatments, with five replicates for each combination. In June 2013, a total of 120 containers (15 × 15 × 15 cm) were filled with one out of the following six substrates: $D_{0\%}$, $D_{20\%}$, $D_{30\%}$, $D_{50\%}$, *S* and *F* (Table 1). The four seed mixture treatments included a *control* treatment in which no seed mixture was added (to examine the "seedbank" in the substrates) and three treatments in which 1 g of a seed mixture was applied. The three seed mixtures used were (1) a "*forb seed mixture*" containing tall forbs and no grasses, (2) a "*diverse seed mixture*" containing mainly low forbs, *Sedum* and some grasses and (3) a "*grass seed mixture*" that was similar to the diverse seed mixture, but also contained seeds of *Festuca rubra*, which is a common and often dominant grass species on green roofs in the surroundings of the study site (see detailed seed mixture compositions in Table S1). Containers were placed randomly in a climate controlled greenhouse with 70% RH, 16 h at 21 °C (day) and 8 h at 16 °C (night). Natural daylight was supplemented by 400 W metal halide lamps (225 μmol s^{-1} m^{-2} photosynthetically active radiation, one lamp per 1.5 m^2). The containers were watered regularly (at least three times per week) and received sufficient water for ten weeks.

2.1.2. Experimental blue-green roof

The experimental blue-green roof is situated on the building of the Netherlands Institute of Ecology (NIOO-KNAW) in Wageningen (51.9866°N, 5.6716°E) and was constructed in 2012. The region has a temperate maritime climate (Köppen Cfb) characterized by cool summers, moderate winters and precipitation throughout the year with an average annual rainfall of 850 mm.

The experimental part of the roof consists of 45 experimental plots of 8 m^2 (2.85 m × 2.85 m). All plots have a 11 cm deep water storage, on top of which a geotextile cloth is placed that holds the substrate. The 45 experimental plots were divided over nine different treatments with five replicates each (Fig. 1; Table 1). The treatments were randomly assigned to the 45 available plots on the roof (Fig. 1). Seed mixtures were sown upon roof construction in 2012. All plots were fully sun- and rain-exposed and did not receive any shading nor irrigation. The only maintenance on the roof was removal of tree saplings once per year in autumn.

Each treatment differed in its combination of substrate composition, substrate depth and seed mixture (Table 1; Supplementary information Table S1). The nine treatments were selected in consultation with industrial green roof designers to create a variety of combinations of seed mixtures and substrates that would meet criteria interesting for market introduction in terms of costs, attractiveness and potential for plant biodiversity. Two of the treatments were controls resembling conventional roof system. Firstly, the *Gravel* treatment resembled a conventional (non-green) flat roof covered with 100% gravel. Secondly, the *Sedum* treatment resembled a conventional extensive green roof, where Sedum plugs were applied to create an immediate *Sedum* cover. Other treatments included four treatments that mainly differed in the ratio between lightweight substrate (red lava and fine pumice) and heavy dense substrate (compost, peat and sand), such that the substrates in treatments consisted for either 0%, 20%, 30% or 50% of dense material (treatments $D_{0\%}$, $D_{20\%}$, $D_{30\%}$, $D_{50\%}$, respectively). Two treatments were characterized by the addition of 40% local soil, which was sandy soil collected from the direct environment of the building. Treatments with local soil were added to the experiment since we were interested in how addition of local soil could benefit biodiversity on blue-green roof systems. Those two *Soil* treatments differed between eachother in their substrate depth, being either 60 mm or 90 mm (treatments *S* and S_9, respectively). The ninth treatment had a substrate that contained only 10% dense materials, but was fertilized (treatment *F*; Table 1). Due to the limited roof space it was not possible to study in-situ all combinations of substrates and initial seed mixture compositions. Consequently, it can sometimes not be differentiated whether differences in vegetation between treatments (e.g. $D_{0\%}$ and $D_{20\%}$) are caused by differences in

Table 1

Overview of substrate compositions and treatments in the experimental setup on the blue-green roof. ‘Dense’ treatments refer to the percentage of dense substrate in the treatment (compost + peat + sand). The substrate compositions of treatments $D_{0\%}$, $D_{20\%}$, $D_{30\%}$, $D_{50\%}$, *S* and *F* were also used in the greenhouse experiment, and applied in combination with seed mixtures (F = forb seed mixture, D = diverse seed mixture, G = grass seed mixture, S = Sedum seed mixture). VWC = Volumetric Water Content in winter, average of the months November to December in the years 2015 and 2017. See Table S1 for the composition of the seed mixtures.

Treatment		Substrate composition (%)						Dense substrate (%)	VWC	Depth (mm)	Seed mixture
		Fractured tiles	Red lava	Fine pumice	Compost	Peat	Other				
Dense 0%	*($D_{0\%}$)*		100					0	0.12	60	F
Dense 20%	*($D_{20\%}$)*		55	25	15	5		20	0.17	60	D
Dense 30%	*($D_{30\%}$)*		45	25	20	10		30	0.28	60	F
Dense 50%	*($D_{50\%}$)*		30	20	20	15	15 sand	50	0.28	60	G
Soil	*(S)*	60					40 local soil	40	0.31	60	D
Soil 9 cm	*(S_9)*	60					40 local soil	40	0.28	90	D
Fertilized	*(F)*		90		10			10	0.18	60	G
Sedum	*(Sed)*	30	30	15	20		5 coconut fibre	25	0.27	60	S
Gravel	*(Grv)*						100 gravel	0	–	60	–

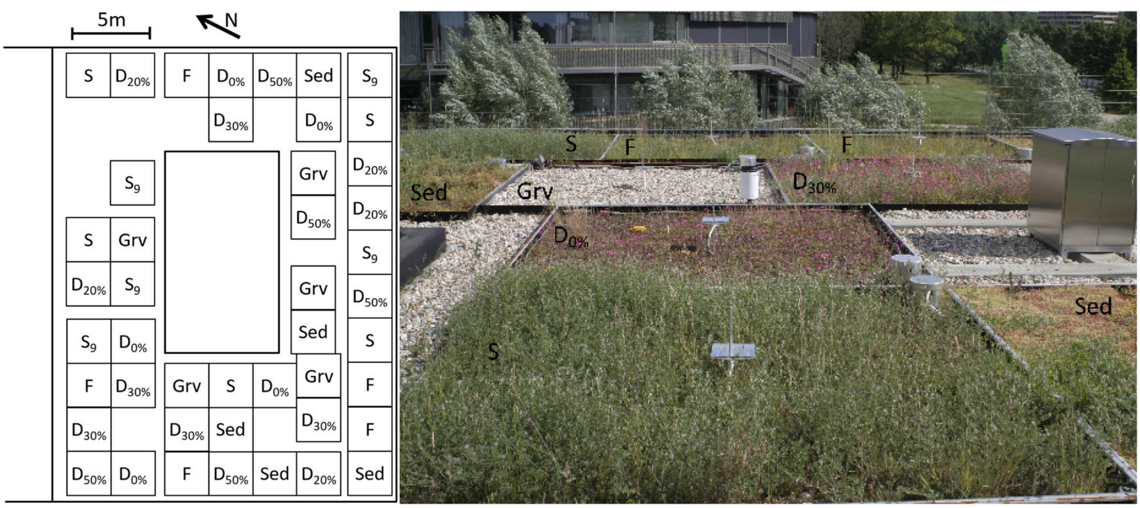

Fig. 1. Experimental setup of the blue-green roof. Treatments: $D_{0\%}$ = Dense 0%, $D_{20\%}$ = Dense 20%, $D_{30\%}$ = Dense 30%, $D_{50\%}$ = Dense 50%, S = Soil, S_9 = Soil 9 cm, F = Fertilized, Sed = Sedum, Grv = Gravel. See Table 1 for more details on the different treatments. Picture of the roof taken on 11 June 2014 by Gerdien Bos-Groenendijk. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

substrate composition or initial seed mixture, which we further comment on in the Discussion.

2.2. Data collection

2.2.1. Greenhouse experiment

Ten weeks after seed mixtures were sown, in each container all plant species were identified and the aboveground biomass was clipped at substrate level. The biomass was sorted on species and the proportion of biomass for each species was estimated. The total harvested biomass was then dried at 70 °C and the dry mass was measured.

2.2.2. Experimental blue-green roof

Vegetation surveys were done annually during nine years following roof construction, except for 2016 in which no survey was done. Vegetation surveys were done on the following dates: 4 July 2013, 14 July 2014, 20 July 2015, 21 July 2017, 30 June 2018, 27 June 2019, 29 June 2020 and 25 June 2021. All plant species and their cover (in %) were recorded in a permanent quadrat of 1 m^2 area (1 × 1 m) in each plot. Permanent quadrats were positioned at least 10 cm from the edge of the plot. We observed and confirmed throughout the study period that the vegetation within a quadrat was representative for the vegetation in a whole plot.

In each plot a Campbell CS616 sensor (30 cm long probe) and an EC-5 sensor (5 cm long probe) were used to measure volumetric water content. Unfortunately, the data was inconsistent for most years due to maintenance on the non-green part of the roof or sensor malfunctioning. We therefore only used data from November–December from 2015 and 2017 to calculate the volumetric water content in winter, i.e. when soils were saturated and the volumetric water content approached the maximum water holding capacity. An average volumetric water content was calculated for each treatment.

Precipitation data were provided by the Royal Netherlands Meteorological Institute from the meteorological station Deelen (52.06°N, 5.89°E), located 17 km northeast of the blue-green roof (KNMI, 2020).

2.3. Data analysis

All data processing and analysis were done in R (R Core Team, 2019).

2.3.1. Greenhouse experiment

An ANOVA was used to test whether seed mixture and substrate significantly explained variation in total biomass and species richness. A post-hoc Tukey test was used to detect significant differences among individual treatments.

2.3.2. Experimental blue-green roof

Firstly, we determined how frequent forb species were recorded on the blue-green roof, which is a useful statistic to optimize seed mixtures that can be applied on new blue-green roofs. For this purpose, we derived the number of plot-years (maximum = 360 plot-years, eight annual surveys of 45 plots) and the number of years in which each species was recorded (maximum = eight years). For all forb species that were included in seed mixtures and were sown after construction of the blue-green roof, we determined whether they were either not recorded at all, occasionally recorded (present in at least one plot in maximum half of the years), or frequently recorded (present in at least one plot in more than half of the years). We also determined which forb species that were not sown were successful, i.e. which plant species spontaneously appeared on the roof and were recorded in at least 30 plot-years. Lastly, we determined which species were recorded in at least 20 plot-years and were mostly confined to treatments with local soil (i.e. more than half of the records were in treatments *S* and S_9).

Secondly, we explored how the vegetation composition changed over the years in each treatment, by performing non-metric multidimensional scaling (NMDS) ordination using the *metaNMDS* function of the *vegan* R package (Oksanen et al., 2013). The input matrix contained plant cover (in %) of all recorded plant species in 360 surveys (i.e. 45 plots in eight annual surveys). Ordination was based on Bray–Curtis dissimilarity and three dimensions (the minimum number of dimensions for which NMDS stress was lower than 0.2).

We focussed the rest of our analysis on six vegetation characteristics: (1) Species richness, (2) Shannon diversity index, (3) Total plant cover (%), (4) Forb cover (%), (5) Grass cover (%) and (6) *Sedum* cover (%). The Shannon diversity index was for each plot calculated as follows (Shannon, 1948):

$$H' = \sum_{i=1}^{s} p_i ln p_i \quad (1)$$

where H' is the diversity index, s is the number of species recorded in the plot, and p_i is the cover of plant species i, expressed as the proportion of the total plant cover (calculated as the sum of the cover of all species recorded in the plot).

For each of the six vegetation characteristics, we visualized temporal changes for all treatments. We then used an ANOVA to test whether average vegetation characteristics significantly differed among treatments. For each vegetation characteristic, the means per plot (i.e. for each plot the mean of eight annual surveys was calculated) were used as response variable and treatment was used as explanatory variable. A post-hoc Tukey test was used to detect significant differences among individual treatments.

A linear mixed model was then used to analyse whether vegetation characteristics were corelated with roof age in years (2012 = year 0), the sum of precipitation in April–June (standardized) and percentage of dense substrate. Volumetric water content was not added as explanatory variable, since it was highly correlated with the percentage of dense substrate ($R^2 = 0.79$, $n = 8$). Plot identity nested in treatment and year were added as random intercepts. Since testing three variables for six vegetation characteristics resulted in 18 different tests, we applied the Benjamini–Hochberg procedure to detect which *P* values were significant using a false discovery rate of 0.05 (Benjamini and Hochberg, 1995).

3. Results

3.1. Greenhouse experiment

Total species richness was significantly higher in containers which received the diverse seed mixture than in containers that received the forb seed mixture (Fig. 2a; Fig. S1). Containers that received the forb seed mixture contained almost exclusively forb species, whereas containers that received the diverse seed mixture or grass seed mixture also contained grass and *Sedum* species. There were no significant differences in species richness among substrates (Fig. 2b; Fig. S1).

Total biomass was significantly higher in containers which received the grass seed mixture than in containers that received the diverse seed mixture. Forb biomass was highest in containers that received the forb seed mixture, whereas grass biomass was high in containers that received the grass seed mixture (Fig. 2c; Fig. S1). There were also significant differences in aboveground biomass among containers with different substrates. Containers in which local soil was applied (substrate *S*) had significantly more biomass than all other substrates, whereas containers containing a substrate consisting of 100% red lava (substrate $D_{0\%}$) had significantly lower biomass than all other treatments (Fig. 2d; Fig. S1). Very little biomass was produced in control containers, which did not receive seeds, indicating that all substrates had no or a limited seedbank.

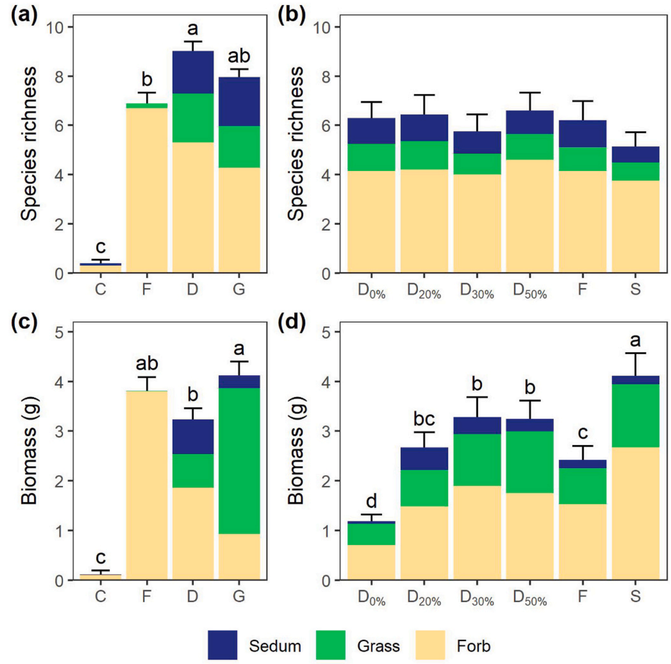

Fig. 2. Species richness and aboveground dry biomass in containers in the greenhouse experiment ten weeks after seed mixtures were sown. Differences in species richness (mean ± SE) and dry aboveground biomass (mean ± SE) among containers with different seed mixture treatments (a, c) and among containers with different substrates (b, d) are shown. (a) and (c) depict means of all substrates, whereas (b) and (d) depict means of all seed mixture treatments. Seed mixture treatments: C = Control, F = Forb seed mixture, D = Diverse seed mixture, G = Grass seed mixture. See Table 1 for explanation of substrate treatments. Letters above bars indicate significant differences within panels in species richness (a) or in total aboveground biomass (c-d). There were no significant differences in species richness among substrates. Fig. S1 shows the results of all combinations of seed mixtures and substrates.

3.2. Experimental blue-green roof

3.2.1. Plant species presence

A total of 131 plant taxa were recorded in the 45 experimental plots during eight surveys in 2013–2021. Not all plant species that were included in the seed mixture were recorded on the experimental blue-green roof (Fig. 3). For example, *Armeria maritima* and *Erigeron acris* were sown in five different treatments, but were never recorded throughout the study period. Other species, for example *Silene vulgaris*, were present in the first few years but disappeared quickly. Species that were sown and successful, i.e. recorded in at least one plot in most surveys, included *Achillea millefolium*, *Allium schoenoprasum*, *Thymus pulegoides* and *Trifolium arvense* (Fig. 3).

Many forb species appeared spontaneously on the blue-green roof and many of them were recorded frequently, including *Cerastium fontanum*, *Hypochaeris radicata* and *Linaria vulgaris*. Three forb species were predominantly recorded in substrates with local soil: *Medicago lupulina*, *Trifolium repens* and *Vicia hirsuta* (Fig. 3).

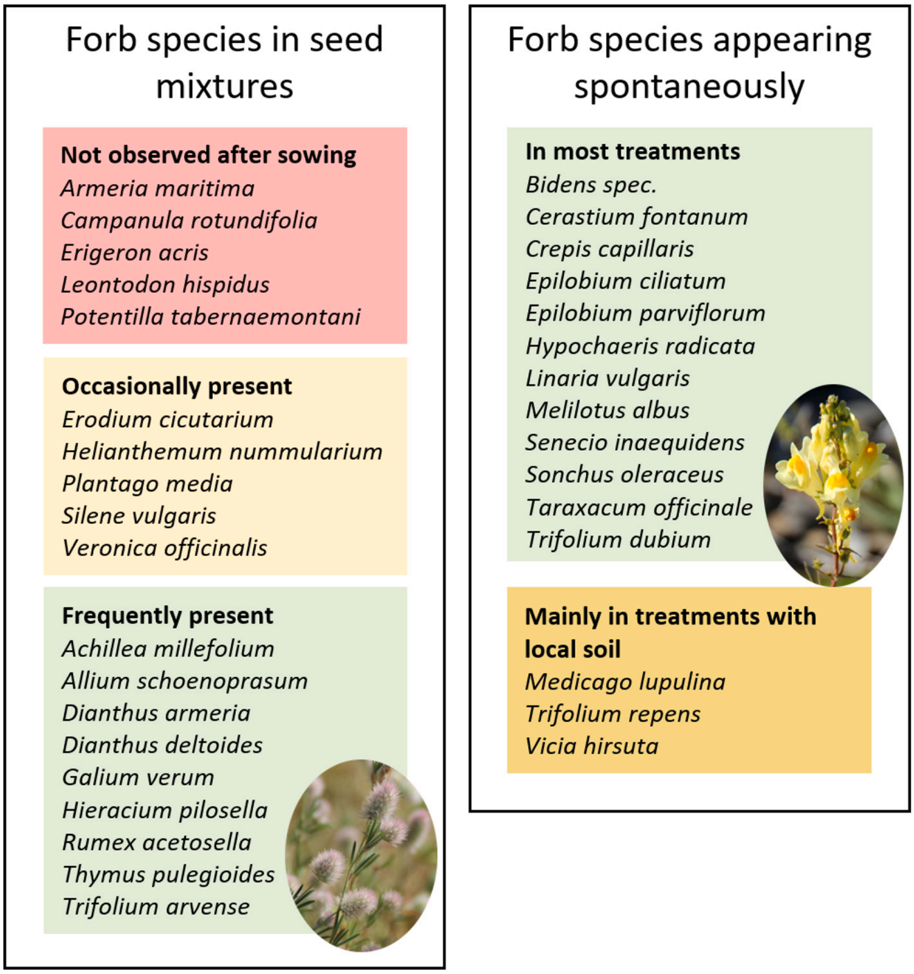

Fig. 3. Forb species on the experimental blue-green roof. The left column shows the fate of sown forb species: 1) not observed at all, 2) occasionally present (maximum observed in four out of eight years), or 3) frequently observed and present in most years. The right column presents a list of the most common forb species that appeared spontaneously on the roof: 1) forb species that were observed in at least 30 plot-years, 2) plant species that were observed in at least 20 plot-years of which more than half in plots with local soil, treatments *S* and S_9). Insets show pictures of *Trifolium arvense* (left column) and *Linaria vulgaris* (right column). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

3.2.2. Vegetation development throughout time

The vegetation composition varied among treatments and changed throughout the study period, which we explored by performing a NMDS ordination (NMDS stress = 0.189, three dimensions, Fig. 4) and by visualizing temporal changes of six different vegetation characteristics (Fig. 5). Plots in most treatments were almost or completely covered by vegetation in 2014, two years after roof construction (Fig. 5c). However, total plant cover never approached 100% in treatments *Gravel*, $D_{0\%}$ and $D_{20\%}$, with cover being lowest in the *Gravel* control treatment (Fig. 5). The scarce vegetation in *Gravel* and $D_{0\%}$ treatments was composed of several forb species: *Linaria vulgaris* and *Bidens frondosa* were often recorded in the *Gravel* treatment, and *Thymus pulegioides* in the $D_{0\%}$ treatment (Fig. 4). The vegetation in most other treatments was in many years dominated by *Festuca* spec. and *Trifolium arvense*. Of all recorded plant cover, 35.6% and 19.8% belonged to those species, respectively. *Trifolium arvense* was abundant in many treatments during the early years of the experiment, resulting in a high total forb cover especially in 2014 and 2015 (Fig. 5d). The cover of *Festuca* spec. Increased from 2015 onwards, which resulted in a high grass cover in many treatments in 2015–2018 (Fig. 4, Fig. 5e). Grass cover decreased again from 2019 onwards, coinciding with an increase in forb and *Sedum* cover (Fig. 5d-f). In 2020, both species richness and total plant cover decreased in almost all treatments. Species richness recovered in all treatments in 2021, but plant cover did not recover in $D_{20\%}$ and $D_{50\%}$ treatments (Fig. 5). The vegetation in the *Sedum* treatment differed from most other treatments: Due to the application of *Sedum* plugs, *Sedum* cover was until 2018 higher than in all other treatments (Fig. 5c).

3.2.3. Factors explaining differences in vegetation characteristics

All six vegetation characteristics that we investigated differed significantly among treatments (Fig. 5, Fig. 6). Plant species richness was highest in treatments where the substrate contained local soil, averaging 11.8 and 11.2 plant species in a 1 m^2 plot for the treatments with substrate heights of 60 mm (treatment *S*) and 90 mm (treatment S_9), respectively. Plant species richness was lowest in plots with gravel, representing conventional non-green flat roofs, averaging only 4.8 species in a 1 m^2 plot (Fig. 6a). There was less variation in the Shannon diversity index among treatments, but the diversity index was significantly lower in treatment $D_{50\%}$ in comparison to most other treatments (Fig. 6b).

Total vegetation cover was lowest in gravel plots and averaged only 8.9%. Total vegetation cover was also low for other treatments where the substrate contained little dense material. Specifically, total cover was one average 21% and 57% for treatments $D_{0\%}$ and $D_{20\%}$, respectively. Total vegetation cover in all other treatments was on average over 75%, and was especially high in the *Sedum* treatment (98%; Fig. 6c). Mean forb cover was highest in $D_{30\%}$ and S_9 treatments (Fig. 6d), whereas grass cover was high in $D_{50\%}$, *S*, S_9 and *F* treatments (Fig. 6e). *Sedum* cover was highest in the *Sedum* treatment (all-year average 45%) and elsewhere most apparent in the $D_{20\%}$ (17%) and S_9 treatments (14%; Fig. 6f).

None of the six vegetation characteristics were significantly correlated with either spring precipitation, roof age or the percentage of dense substrate (Table 2).

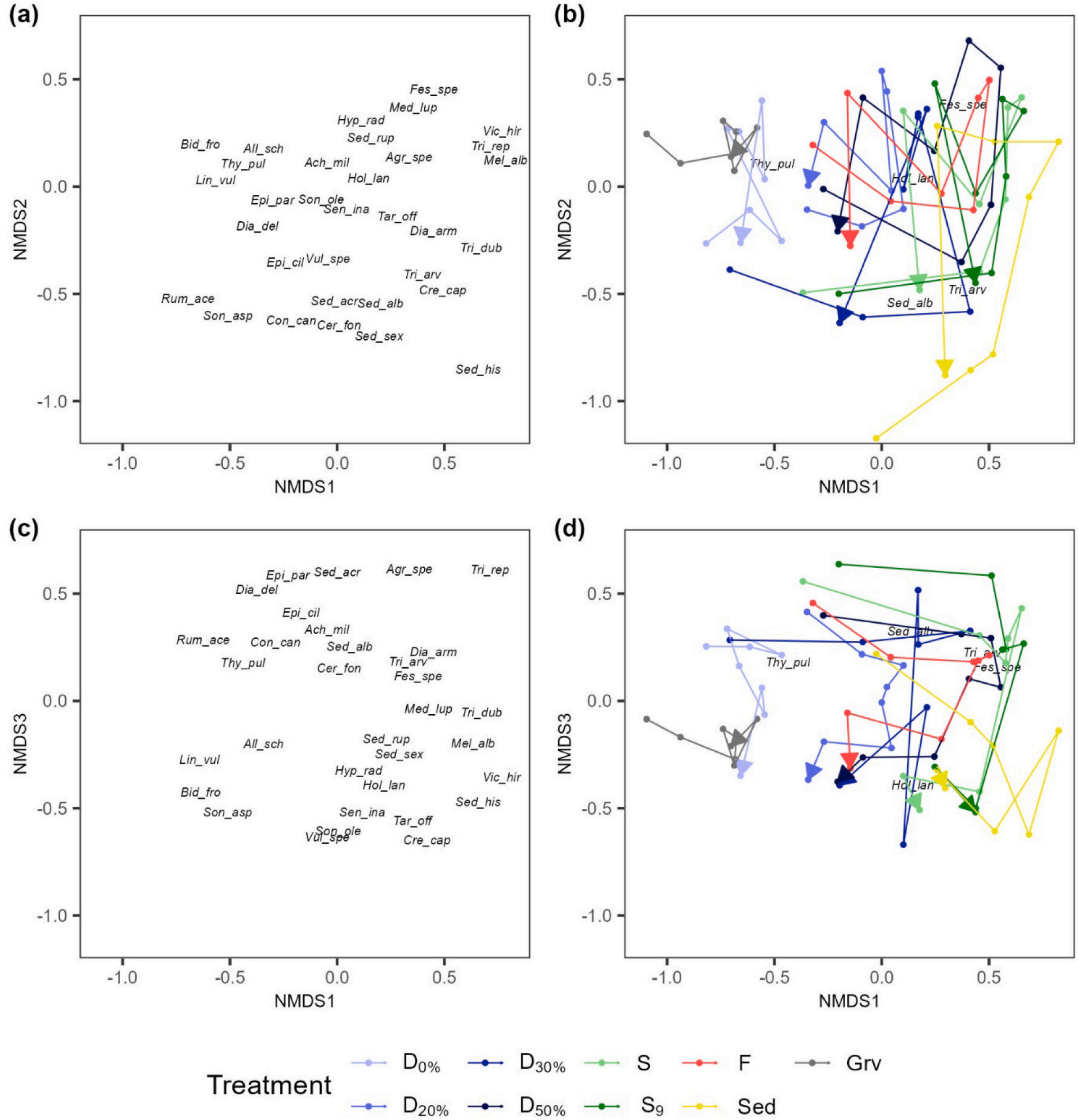

Fig. 4. NMDS analysis showing temporal shifts in vegetation composition for each treatment. Species positions and shifts in vegetation composition are shown on the first and second NMDS axis in (a-b) and on the first and third axis in (c-d). Lines in (b) and (d) show vegetation shifts of each treatment during the years 2013–2021 (excluding 2016, when no survey was done), where each point represents the mean of a treatment in one year and where the arrows heads point at the last survey year 2021. All species were included in the analysis, but in (a) and (c) only plant species are shown that were recorded at least 25 times. In (b) and (d) a selection of five common species is shown for visual guidance. NMDS stress = 0.189. Plant name abbreviations: Ach_mil = *Achillea millefolium*, Agr_spe = *Agrostis spec.*, All_sch = *Allium schoenoprasum*, Bid_fro = *Bidens frondosa*, Cer_fon = *Cerastium fontanum*, Con_can = *Conyza canadensis*, Cre_cap = *Crepis capillaris*, Dia_arm = *Dianthus armeria*, Dia_del = *Dianthus deltoides*, Epi_cil = *Epilobium ciliatum*, Epi_par = *Epilobium parviflorum*, Fes_spe = *Festuca* spec., Hol_lan = *Holcus lanatus*, Hyp_rad = *Hypochaeris radicata*, Lin_vul = *Linaria vulgaris*, Med_lup = *Medicago lupulina*, Mel_alb = *Melilotus albus*, Rum_ace = *Rumex acetosella*, Sed_acr = *Sedum acre*, Sed_alb = *Sedum album*, Sed_his = *Sedum hispanicum*, Sed_rup = *Sedum rupestre*, Sed_sex = *Sedum sexangulare*, Sen_ina = *Senecio inaequidens*, Son_asp = *Sonchus asper*, Son_ole = *Sonchus oleraceus*, Tar_off = *Taraxacum officinale*, Thy_pul = *Thymus pulegioides*, Tri_arv = *Trifolium arvense*, Tri_dub = *Trifolium dubium*, Tri_rep = *Trifolium repens*, Vic_hir = *Vicia hirsuta*, Vul_spe = *Vulpia* spec.

4. Discussion

We studied the vegetation development for nine different treatments in an experimental setting on a blue-green roof. Two of the treatments resembled conventional roof systems: A conventional non-green gravel roof and a conventional *Sedum* green roof. The other treatments were designed with different combinations of substrate composition, substrate depth and seed mixture sown upon roof construction. It is evident that on all green roof substrates plant species richness and often also plant cover was higher than on the gravel substrate, providing further evidence for the hypothesis that green roofs support a higher biodiversity than conventional roofs (Williams et al., 2014).

4.1. *Differences in vegetation characteristics among blue-green roof designs*

The treatments $D_{0\%}$, $D_{20\%}$, *Gravel* and *Sedum* showed unique vegetation characteristics, whereas the other five treatments ($D_{30\%}$, $D_{50\%}$, *S*,

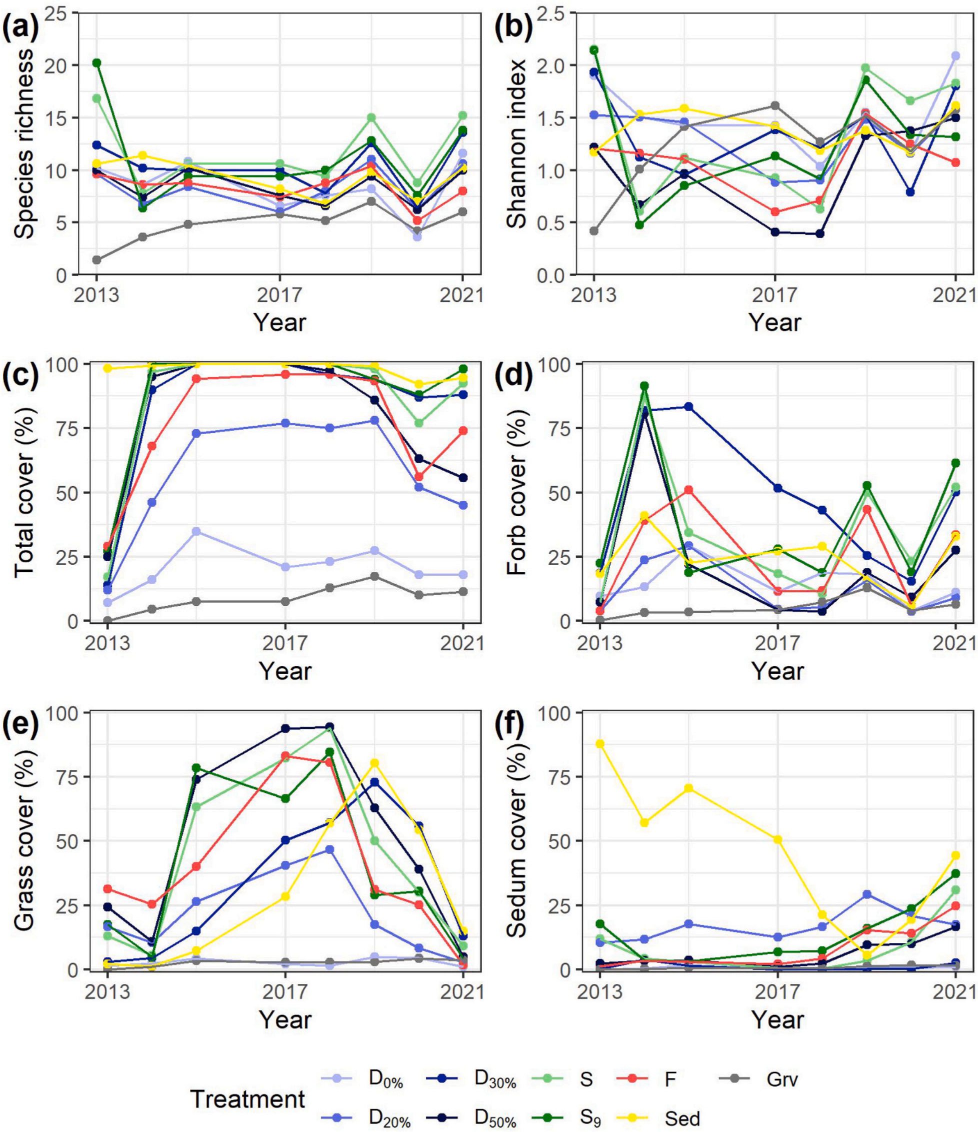

Fig. 5. Development of (a) mean species richness, (b) Shannon index, (c) total cover, (d) forb cover, (e) grass cover and (f) Sedum cover throughout the years in each treatment. No survey was done in 2016.

S_9, F) were more similar to each other. The vegetation in $D_{0\%}$, $D_{20\%}$ and *Gravel* treatments always remained at least somewhat open, likely because the lack of nutrients in the substrate prevented the vegetation to become completely closed. In all other treatments, the vegetation closed within 2–3 year after roof construction. When aiming for a closed vegetation, our results suggests that the (unfertilized) substrate should contain at least 30% organic content. An excessive amount of organic material may, however, result in the dominance of grasses, and 30–40% dense material in the substrate seems to be an optimum among our treatments where species richness and forb cover are maximized long-term. Increasing the amount of organic materials in the substrate comes at the costs of increased substrate weight and the potential to leach nutrients via runoff water (Rowe et al., 2006; Lata et al., 2018).

We acknowledge that our experimental blue-green roof design is not an optimal factorial experimental design. Differences between some plots (e.g. $D_{0\%}$ and $D_{20\%}$) may have been caused by either substrate composition or initial seed mixture. The greenhouse experiment revealed that the applied seed mixture has a large impact on short-term species richness and aboveground biomass in containers in standardized conditions. Also on the blue-green roof the seed mixture may initially be an important determinant of species richness and species composition. However, throughout the years the relative impact of the application of the seed mixtures may fade and substrate composition may have a larger role in shaping the vegetation. For example, species richness did not differ among substrates in the greenhouse experiment, but species richness was significantly higher in treatments S and S_9 than in treatment $D_{20\%}$ throughout the long-term blue-green roof experiment, despite the same seed mixture that was applied. Similarly, grasses developed on the blue-green roof even in treatments where the forb seed mixture was applied. For example, grass cover was not significantly lower in one of the treatments where the forb seed mixture was applied (treatment $D_{30\%}$) compared to one of the treatments where the grass seed mixture was applied (treatment F). We thus suggest that seed mixture may be important for initial vegetation development, but that substrate composition is more important for vegetation development long-term.

The average species richness was highest in the treatments where local soil was added to the substrate and in those treatments especially high in the first year (Fig. 5). Unfortunately, we cannot disentangle the relative importance of the addition of local soil and the application of the diverse seed mixture on the effect on species richness. In the short-term greenhouse experiment, application of the diverse seed mixture

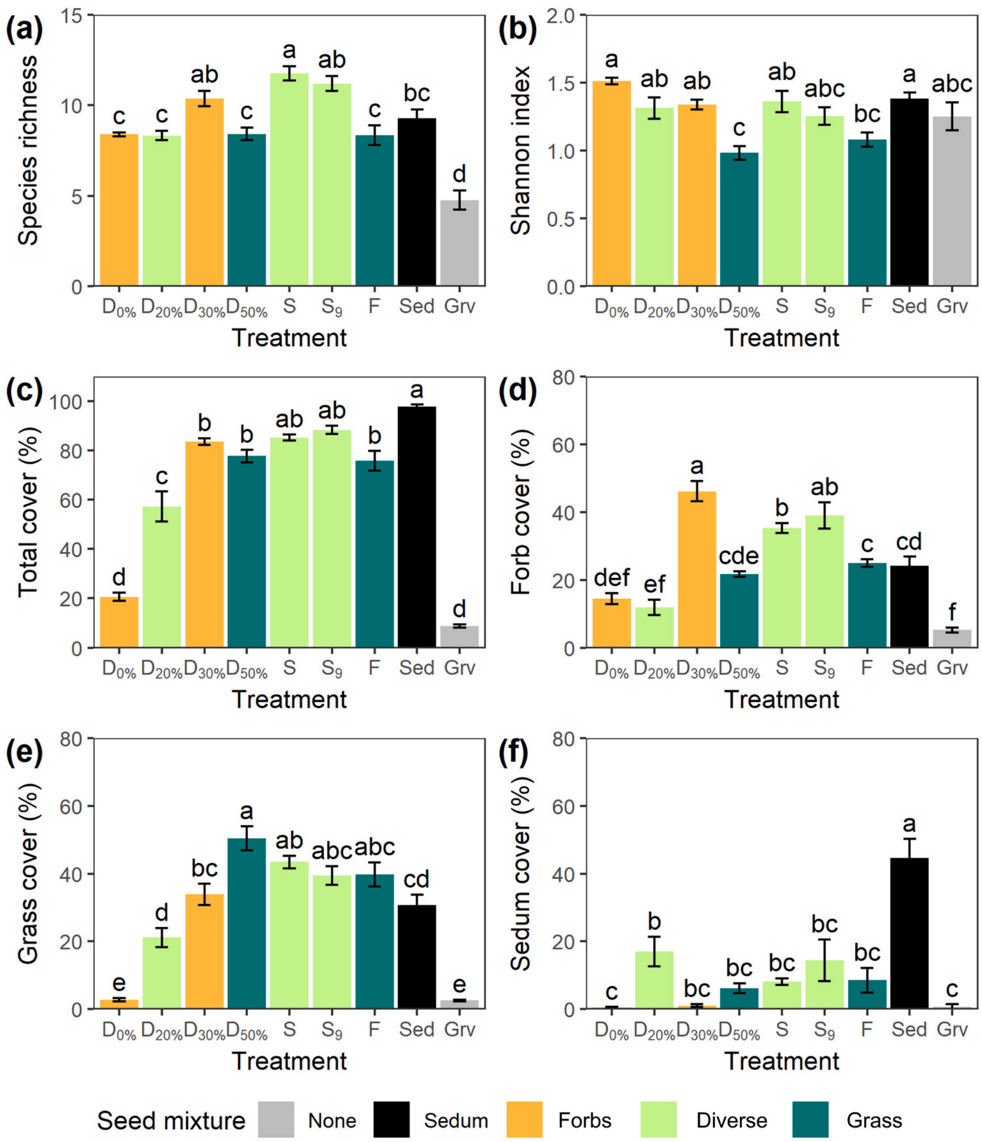

Fig. 6. Differences in (a) mean species richness, (b) Shannon index, (c) total cover, (d) forb cover, (e) grass cover and (f) *Sedum* cover among treatments throughout the whole study period. Values were averaged per plot before the analysis (five plots for each of the nine treatments, n = 45 plots). Error bars are standard errors. Letters indicate significant differences among treatments.

Table 2
Linear mixed model results for the effects of precipitation (standardized sum of precipitation in April–June), roof age (2012 = year 0) and percentage of dense substrate on six vegetation characteristics. None of the *P* values were significant following the Benjamini-Hochberg procedure.

	Species richness			Shannon index			Total cover (%)		
	Est.	SE	P	Est.	SE	P	Est.	SE	P
Intercept	7.72	1.75		1.27	0.23		28.29	18.98	
Precipitation	0.75	0.70	0.334	0.07	0.10	0.529	−0.95	6.73	0.893
Roof age	−0.11	0.26	0.676	0.02	0.04	0.678	0.62	2.48	0.812
Dense substrate (%)	0.08	0.03	0.045	0.00	0.00	0.324	1.43	0.43	0.012
	Forb cover (%)			Grass cover (%)			Sedum cover (%)		
	Est.	SE	P	Est.	SE	P	Est.	SE	P
Intercept	21.04	10.64		−0.40	13.23		7.49	8.80	
Precipitation	9.41	4.00	0.065	−12.91	5.70	0.073	2.63	1.33	0.105
Roof age	−1.45	1.47	0.370	1.97	2.10	0.391	0.08	0.49	0.872
Dense substrate (%)	0.47	0.22	0.071	0.82	0.18	0.003	0.14	0.29	0.638

resulted in the highest species richness, whereas substrate composition did not affect species richness. However, some forb species that were not sown were almost exclusively recorded in treatments where local soil was applied, showing how the application of local soil may boost plant diversity and stimulate native plant species.

Despite substrate depth being one of the main determinants of plant growth on green roofs (e.g. Durhman et al., 2007; Rowe et al., 2012; Vandegrift et al., 2019), in our study there was no significant difference in any of the vegetation characteristics between the two treatments where local soil was applied that differed in substrate depth. One of the reasons why increased substrate depth promotes plant diversity is that it holds more water and reduces drought stress (Brown and Lundholm, 2015), but this feature may become less important on blue-green roofs where the water storage underneath reduces drought stress through evaporation. Future experiments including treatments with and without water storage in combination with different substrate depths can quantify the relative importance of substrate depth and water storage on the vegetation.

4.2. Weather influences on blue-green roof vegetation

Weather has a large influence on green roof vegetations (Köhler, 2006; Bates et al., 2013). Previous studies have shown how non-succulent plants thrive better on roofs at locations with more precipitation in Scandinavia (Lönnqvist et al., 2021). In 2020 we observed starvation among grasses and some abundant forbs (e.g. *Trifolium arvense*) which flourished in earlier years, possibly because of the extremely dry conditions (the spring of 2020 was the driest in the study period). The starvation of grasses created open space that may have had a positive influence on species richness in 2021. However, none of the vegetation variables was significantly associated with rainfall in spring. This is not necessarily surprising, since long time series are needed to analyse effects of weather. Even though our study is relatively long-term and contains eight years of data, a longer time series is probably needed to quantify effects of weather on blue-green roof vegetation in more detail. Natural succession may determine vegetation development especially in the first years, which further complicates analysis of weather variables. For example, on our roof the vegetation was one year after roof construction still very open and only closed in most treatments in the second year after roof construction.

4.3. Blue-green versus green roof vegetation

Our experimental roof is a blue-green roof that has an 11 cm deep water storage compartment under the substrate layer, opposed to conventional green roof systems that lack such an additional water storage compartment. This implies that the vegetation on blue-green roofs may experience less drought stress, which is generally a limiting factor for plant growth (Durhman et al., 2006), and may therefore support growth of a higher diversity of (non-succulent) plants species. This study provided an excellent opportunity to compare plant species richness and composition between a blue-green roof and a conventional green roof that is situated on the same building and was studied over the same time period (Van der Kolk et al., 2020). Our experimental blue-green treatments occupy one half of the roof, whereas the other half is an extensive green roof constructed in 2020 with substrate depths varying between 6 and 20 cm and without additional water storage capacity. The species richness on this adjacent green roof was on average 7.1 species on a sun-exposed 1 m^2 plot with a substrate depth of 6 cm between 2012 and 2019 (Van der Kolk et al., 2020). The *Sedum* treatment on the blue-green experimental roof has a similar substrate design, but supported on average 9.3 species on a 1 m^2 plot. Another noticeable difference between the roofs is that *Sedum* cover in the *Sedum* treatment on the blue-green roof dramatically decreased throughout the study period, probably because forbs and grasses outcompeted the succulents. In contrast, on the adjacent green roof *Sedum* cover was high throughout the whole study period (Van der Kolk et al., 2020). These observations suggest that the multifunctional design of blue-green roofs also benefits plant species richness. Another example of how green roof vegetations can benefit from multifunctional designs are solar panels placed at a height of 1 m above an extensive (non-blue) green roof surface, which by creating shade can reduce drought stress and stimulate plant diversity (Van der Kolk and van den Berg, 2019).

4.4. Conclusions and recommendations

In conclusion, we show that on blue-green roof systems the composition of the initially applied seed mixture is important for vegetation development and plant species richness short-term, whereas the substrate composition is more important long-term. For the construction of new blue-green roof systems in our (climate) region, we recommend to use a substrate with a depth of 60 mm that contains 30% dense organic material, preferably locally collected soil to boost growth of native plant species, and 70% light-weight material (e.g. pumice).

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.

Data availability

Data is available online at https://doi.org/10.5281/zenodo.7307456.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecoleng.2022.106847.

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