#### **ORIGINAL PAPER**



# A main effects meta principal components analysis of netting effects on fruit: using apple as a model crop

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#### Abstract

To differentiate effects of netting and attribute them to crop, cultivar, planting density, climate, net type and colour, ca. 200 publications were scanned originally. Apple was chosen as a model crop due to the majority of reports, wide variation with many varieties and growing locations worldwide in the Northern and Southern hemisphere, but the results may be useful for other fruiting plants. After meeting strict selection criteria, a meta-analysis of 26 internationally published peer-reviewed articles was based on seven varieties and seventeen locations with a diverse range of climates. A novel Main Effects Meta Principal Components Analysis (ME Meta-PCA) was developed and provided unexpectedly uniform results: Location (climate), planting density and hail net (type and colour) had negligible impacts. Fruit (red) colour, most adversely affected by netting, correlated with TSS viz fruit sweetness, as often postulated in consumer studies, followed, to a smaller extent, by sugar/TSS, fruit firmness and acidity but small increase in fruit mass—i.e. maintenance of fruit quality under netting over all seven varieties (Braeburn, Gala, Elstar, Jonagold, Pinova and Fuji) examined and locations worldwide. While Jonagold and the early ripening Gala appeared suitable, unaffected and stable in the netting effects in the ME Meta-PCA, Pinova was the least suitable for cultivation under netting. Interestingly, late ripening cultivars (Braeburn and Cripps Pink) were both positively influenced by desired earlier ripening under netting. These effects on fruit quality are discussed with respect to shade adaptation under netting and countermeasures such as easy colouring mutants or reflective mulches.

**Keywords** Climate change  $\cdot$  Fruit colour  $\cdot$  Fruit quality  $\cdot$  Hail net  $\cdot$  ME Meta Principal component analysis (ME Meta-PCA)  $\cdot$  Shade

# Introduction

As a result of climate change, plants are often covered with netting to protect them from different environmental influences. When plants are grown in an open field, they are increasingly exposed to hail storms, excessive solar irradiation and pests. Hail netting installed over the tree canopies will beside hailstorm protection also shield the crops from solar irradiation (McCaskill et al. 2016) and will act as a major mechanical obstacle for most pests, thus providing an

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<sup>3</sup> INRES - Horticultural Science, University of Bonn, Bonn, Germany effective infestation barrier e.g. against *Drosophila suzukii*. It effectively creates a separate ecosystem. With anticipated climate change, application of netting will become increasingly important and widespread (Bisbis et al. 2018; Webb et al. 2017).

The adverse effects of nets include interception of solar radiation viz light deprivation for plant growth. This is of particular importance in intensive crop cultivation, where nutrients and water are supplied to the optimum, hence management of light becomes the limiting factor in terms of both fruit quality and quantity (yield) (Demestihas et al. 2017; Robinson et al. 2013). Netting also alters the microclimate inside the orchard (McCaskill et al. 2016; Solomakhin and Blanke 2010) in that it reduces temperature, decreases evapotranspiration and wind speed, while humidity in the orchard increases (Iglesias and Alegre 2006; Middleton and McWaters 2002; Solomakhin and Blanke 2007). This particular ecosystem in turn triggers changes in physiological

processes of affected plants to stabilize their functioning in an altered environment (Zeller et al. 2009).

Published scientific research on the effects of netting focused on specific cultivars and as such provided pieces of information necessary to determine the effects that netting has on individual characteristics of fruit of apple trees. These effects are important from both the physiological and commercial standpoints. The reports range from botanical approaches (Solomakhin and Blanke 2010), via horticultural (Stampar et al. 2001), environmental (Amarante et al. 2009), sustainability, plant hormone signalling, and modelling (Webb et al. 2017) to meteorological papers (McCaskill et al. 2016). Netting protection is seen as a prerequisite for a sustainable future of fruit growing worldwide; the importance of the issue lies in the necessity of providing consistently high fruit quality.

In view of apparent contradictions reported so far, this paper provides an interdisciplinary meta-analytical approach about the impact of netting on fruit characteristics using apple as a model crop; the results may be transferable to other fruit crops. Meta-analysis is a statistically and biometrically sound methodology extensively used in natural sciences (e.g. Hedges and Olkin 1985; Hedges et al. 1999), was adjusted by combining meta-analysis and principal component analysis into main effect meta-PCA (ME Meta-PCA) analysis. This paper covers seven widely-grown apple cultivars: Braeburn, Cripps Pink, Fuji, Gala, Elstar, Jonagold and Pinova with a meta-analysis for the following characteristics: fruit weight, fruit firmness, sugar content, acidity, and starch content. Beside the individual meta-analysis of the effect of netting on each of these measured fruit characteristics in general and across cultivars, all the fruit characteristics and cultivars were analysed in combination through the ME Meta-PCA. The objective of the work was to provide a general yet comprehensive overview of the complex issue of the netting effect in apple. Such Meta-PCA analysis provides a unique overview into grouping of characteristics and enables better insight into physiological mechanisms.

# **Materials and methods**

# **Data collection**

We searched for published peer-reviewed research in academic databases searching for publications on netting effects in apple. Search terms included 'apple', 'netting', 'hail' and 'shade' with Boolean operators and replacement characters. The initial ca. 200 publications were screened on the basis of the following selection criteria: (a) there had to be apple trees at the same site both covered with netting and an uncovered control, (b) there had to be measures of studied characteristics in both treatment and control trees, and (c) additional information about cultivar, netting type and location was available-only 26 papers satisfied these selection criteria. We investigated effects of netting on the following characteristics: fruit firmness, fruit weight, fruit colour, soluble solids, acidity and starch conversion. Recorded were primarily means, variation measures and sample sizes for both treatment and the control. The locations ranged from Santa Catarina and Vacaria (Brazil), Rio Negro (Argentina), Nuble (Chile), Stellenbosch (RSA), Tasmania (Australia), Mollerussa (Spain), Mirosan and Maribor (Slovenia), Falicetto and Bolzano (Italy), Dabrowice (Poland), Wädenswil (Switzerland), Zadar (Croatia), Gradacac (BiH), to Bodensee and Bonn (Germany), covering BSk, Cfa, Cfb, Csa, Csb, Dfa, Dfb climates and included the following apple cultivars: Braeburn, Cripps Pink, Fuji, Gala, Elstar, Jonagold and Pinova. For each study, the following additional information was also recorded: location, netting characteristics, planting density, climatic conditions (Table 1).

Careful analysis of the selected publications provided 11,277 individual data recordings from 537 observations of concomitant comparisons of both netted trees and the control (adjacent trees without hail net) regarding the studied characteristics in the target cultivars. For each of those observations, 21 recordings were made about the means, sample size and standard deviations for both the treatment and the control together with data regarding cultivar, netting characteristics, planting density and climatic conditions. This formed a total data matrix of  $537 \times 21$ , i.e. 11,277 individual data recordings.

In cases, where variation was reported through measures other than standard deviation (SD), it was back calculated. For studies, where no measure of variation was reported, it was calculated on the basis of the overall coefficient of variation, in order to obtain SDs for all observations (Bai et al. 2013; Fu et al. 2013; van Groenigen et al. 2014).

#### **Definitions and data analysis**

All measured characteristics were standardised in one unit per characteristic. Fruit firmness was recorded as kilogram per square centimetre [kg cm<sup>-2</sup>], fruit weight as grams [g], soluble solids as [°Brix], acidity as percentage of malic acid, colour as hue angle and starch conversion as index [1–10]. For the presentation of the magnitude (Friedrich et al. 2008) of netting effect, we used the natural logarithm of the response ratio R (Hedges et al. 1999):

$$\ln R = \ln \left(\frac{M_t}{M_c}\right)$$

where  $M_t$  is the average value of individual fruit characteristic on trees grown under netting and  $M_c$  is the individual value of the apple fruit characteristic from control

 
 Table 1
 Worldwide locations of the trial sites, latitude and their climate

Location	Country	Latitude	Climate	Hail netting	Planting density <sup>a</sup>		
Bonn	Germany	50.5°N	Cfb	Bright, dark	High		
Bodensee	Germany	47.5°N	Csb	Bright, dark	High		
Dabrowice	Poland	52.3°N	Dfb	Bright, dark	High		
Wädenswil	Switzerland	47.2°N	Dfb	Bright, dark	High		
Falicetto	Italy	44.6°N	Cfa	Bright, dark	High		
Bolzano	Italy	46.2°N	Dfb	Dark	High		
Mollerusa	Spain	41.6°N	Cfa	Dark	Medium		
Mirosan	Slovenia	46.2°N	Dfb	Dark	High		
Maribor	Slovenia	46.5°N	Dfb	Dark	High		
Gradacac	BIH	44.8°N	Dfa	Dark	High		
Zadar	Croatia	44.2°N	Csa	Bright, dark	High		
Southern hemisphere							
Nuble	Chile	36.4°S	Csa	Bright, dark	High		
Rio Negro	Argentina	38.9°S	BSk	Bright, dark	Medium		
Vacaria	Brazil	28.3°S	Cfb	Bright, dark	High		
Santa Catarina	Brazil	28.1°S	Cfa	Bright	Low		
Stellenbosch	South Africa	33.9°S	Cfa	Dark	High		
Tasmania	Australia	32.1°S	Cfb	Bright, dark	Low		

<sup>a</sup>High planting density orchards in excess of 2000 (trees  $ha^{-1}$ ), medium 1000–2000 (trees  $ha^{-1}$ ), and low under 1000 (trees  $ha^{-1}$ )

trees without netting. After the meta-analysis and for a better presentation and overview, the mean effect size was back-transformed:

# $e^{\ln R} - 1$

and as such easily observed as percentage change regarding each of the observed characteristics (Bai et al. 2013; van Groenigen et al. 2014; Pittelkow et al. 2014; Qin et al. 2015).

Considering the difference between the studies and observations within the studies, we conducted in R random effects meta-analysis:

# $\hat{\delta} = \delta + \vartheta_i + \varepsilon_i$

where  $\hat{\delta}$  is effect of hail netting on fruit firmness in lnR;  $\delta$  is general effect of netting on fruit firmness;  $\vartheta_i$  is effect of individual studies; and  $\varepsilon_i$  is pooled error. This is standard description of random effects meta-analysis model similar to the fixed-effects model (Chen and Peace 2013) where additional variability component is included representing random variation between the observations ( $\nu_i$ ) distributed as  $\vartheta_i N(0, \tau^2)$ . Hence  $\hat{\delta}_i N(0, \sigma_i^2 + \tau^2)$ , where  $\hat{\tau}^2$  is estimated trough standard Cochran-DerSimonian–Laird procedure (Chen and Peace 2013; Schwarzer et al. 2015).

For the meta-analysis, the significance of the results was established at the 95% confidence interval not overlapping with zero (Broberg et al. 2017; Curtis and Wang 1998). The standard weighting method of inversed variance was used for weighting means in the meta-analysis (van Groenigen et al. 2014; Hedges and Olkin 1985; Schwarzer et al. 2015). Data were checked for publication bias by graphical means i.e. the funnel plot and by formal tests (Chen and Peace 2013; Schwarzer et al. 2015). The data were meta-analysed by using R package "meta" (Chen and Peace 2013; Schwarzer et al. 2015).

Prior to the meta-analysis, we investigated the influence of individual recorded factors such as cultivar, climate, netting characteristics and planting density by fitting mixedmodel in R package nlme (Coutinho et al. 2018; Qin et al. 2015):

$$Y = \mu + \alpha_i + \beta_i + \gamma_k + \delta_l + \rho_m + \epsilon$$

where Y is the dependent variable (individual response characteristics as treatment effect in lnR);  $\mu$  the general intercept;  $\alpha_i$  the cultivar effect (fixed);  $\beta_j$  the effect of bright or dark netting (fixed);  $\gamma_k$  the effect of climate i.e. temperate and warm (fixed);  $\delta_1$  the effect of high or low planting density (fixed);  $\rho_m$  the study effect (random, without specific study in focus); e is the pooled error. In cases where it was possible with regard to data characteristics interactions between variables were also included in the model and checked; significant results were established at p < 0.05.

#### Main effects meta-PCA model

The extracted main effects were analysed in combination through principal components analysis in order to study the overall effects of netting on the studied characteristics across all varieties. This novel approach of using main effects obtained from a meta-analysis in the analysis of principal components is named main effects meta principal components analysis or ME Meta-PCA and was developed during the process of the PhD of the first author (Bosancic et al. 2017).

Principal component analysis is in essence fitting of a low-dimensional subspace to the multivariate high-dimensional dataset and therefore it is one of the most useful tools for data modelling and visualisation (Vidal et al. 2016). Principal components (PC) are in fact directions in which data variability is most spread and which therefore capture most of the information contents in the data (Kong et al. 2017). Principal components analysis historically is used to estimate principal components of a multivariate random variable x (Hotelling 1933; Pearson 1901). The origins of this method are often related to agricultural practices in terms of singular value decomposition in form that resembles PCA (Fisher and Mackenzie 1923). It remains one of the best tools for analysing multivariate datasets in applied biological sciences (Borcard et al. 2011; Iezzoni and Pritts 1991; Legendre and Legendre 1988; Peres-Neto et al. 2003), including multiple characteristics analysis across multiple cultivars in apple (Stanivuković et al. 2017).

Common definition of PCA assumes that if there is a given set of points  $\{x_j\}_{j=1}^N$  which can be also represented as multivariate random variable x which are in dimensions  $R^D$  that we seek to find a subspace of  $S \in R^D$  of dimensions d (where d < D) which will best fit given set of data-points (Vidal et al. 2016). In that subspace every data point  $x_j \in S$  is possible to present as

 $x_i = \mu + Uy_i$ 

where j = 1, 2, ..., N, and  $\mu \in S$  is a point in the new subspace, U is a D × d matrix forming basis for the subspace by its columns, and  $y_j \in \mathbb{R}^D$  is than the vector for the newly formed coordinates of  $x_i$  in the new subspace (Vidal et al. 2016).

However, in the above case the precise representation of the points allows ambiguous arbitrary choices which cause translational ambiguity and change of basis ambiguity (Vidal et al. 2016). This is solved by setting the means of the variable to zero

$$\frac{1}{N}\sum_{y=1}^N y_j = 0$$

and by requiring the columns to be orthonormal, i.e. the columns of U are not orthonormal, which is required in order to solve the ambiguity related to change of basis. By setting to orthonormal  $U^{T}U$  each point in practice will contain some error, or noise, and can be described as

 $x_j = \mu + Uy_j + \varepsilon_j$ 

This is the final practical PCA model describing desired principal component subspace, which contains and practically describes all points by minimising the sum of squared errors.

Vectors in this study represent individual fruit characteristics and objects are the apple varieties under examination. Angles between the vectors indicate correlations between the studied characteristics in terms of netting effect. Characteristics, which are impacted in a similar manner, will be closely represented in the figure. Opposing directions indicate negative correlations. Conversely, orthogonality between the characteristics vectors indicates lack of correlation. Vector length indicates intensity of impact that the treatment had on each particular characteristic. Hence, grouping of vectors indicates similar response regarding the characteristics those vectors represent.

Apple cultivars as objects are placed in close relation to those fruit characteristics' vectors, which are altered to the largest extent due to the netting influence or impact on the particular variety. Position of cultivars in relation to the centre of the plot indicates the sensitivity of each particular variety to the netting effect. Hence grouping of cultivars along lines of vector or correlated vectors indicates increased sensitivity of those varieties to the netting effect on the characteristics, which are represented by the vectors. Grouping of cultivars around centre of the plot indicates increased stability of those varieties in terms of netting effect on studied fruit characteristics.

# Results

Data analysis with the mixed-model showed that cultivar was the principal variable significantly sub-setting the data across measured characteristics. Therefore, subsets were meta-analysed on the basis of the seven studied cultivars. The general overview of the large number of data across multiple characteristics in multiple cultivars is presented through ME Meta-PCA. Overall, the ME Meta-PCA provided unexpectedly uniform and clear results on the effects of netting on fruit colour, starch breakdown, fruit firmness, sugar (TSS) and acidity.

#### Fruit firmness

Netting had a generally small and negative though statistically highly significant (p < 0.001) effect on fruit firmness of -0.019 (with 95% CI between -0.031 and -0.009), i.e. fruit under netting were somewhat less firm than those grown outside in an open field. A breakdown by variety showed a significant (p < 0.039) negative effect on fruit firmness in Cripps Pink (-0.026, with 95% CI -0.0339 to -0.0180) and Elstar (-0.023, with 95% CI - 0.037 to -0.009) and highly significant (p < 0.001) effect in Pinova (-0.186, with 95% CI - 0.226; -0.147) (Fig. 1) and the first indication of cv. 'Pinova' as unsuitable for cultivation under netting, irrespective of planting density and growing region.

# Fruit weight

In general, there was a *statistically significant* (p=0.0339) but *small positive* effect of 0.023 (with 95% CI from 0.007 to 0.041) on fruit weight. There was also a statistically highly significant effect across all studied cultivars. In Cripps Pink, there was a statistically significant (p=0.043) positive effect of 0.042 (95% CI 0.001–0.083). A statistically significant (p=0.012) positive effect of 0.035 (95% CI 0.008–0.062) on fruit weight was also observed for apple cultivar Fuji. A highly significant (p<0.001) positive effect of 0.131 (95% CI 0.101–0.161) was also observed for cultivar Elstar (Fig. 1), possibly due to their larger sensitivity to alternate bearing.

#### **Soluble solids**

The impact of netting on the total soluble solids (TSS, sugar) content in apple fruit was generally negative and small -0.038 (95% CI-0.047 to -0.029), but statistically highly significant (p<0.001). There is also a statistically significantly (p<0.001) different degree of the same impact of netting between studied cultivars. Netting had a statistically highly significant (p<0.001) negative impact on Braeburn -0.029 (95% CI-0.031 to -0.027), Cripps Pink-0.037 (95% CI-0.043 to -0.032), Fuji-0.033 (95% CI-0.035 to -0.031), Gala-0.032 (95% CI-0.033 to -0.030) and Pinova-0.129 (95% CI-0.143; -0.116) (Fig. 1), the second reason why Pinova appears unsuitable for cultivation under netting across all latitudes.

## **Total acidity**

Although the overall effect of hail netting on total acidity (TA) was *not statistically significant* (p=0.222), there is a statistically highly significant (p<0.001) difference in impact on studied cultivars (Fig. 2). In Elstar, there was a

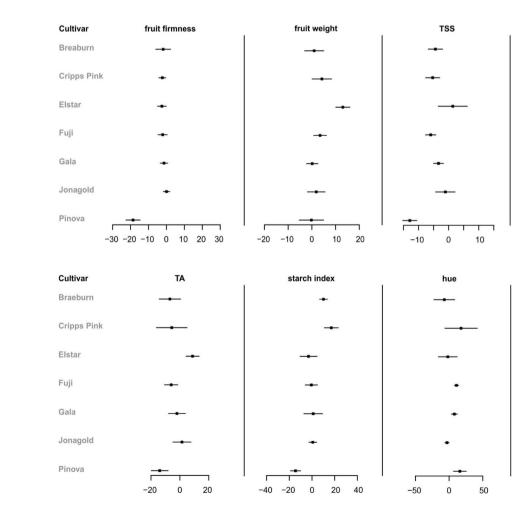


Fig. 1 Forest plots of netting effect size (%) on fruit firmness, fruit weight and total soluble solids (TSS) across cultivars in major fruit growing regions worldwide; error bars are 95% confidence intervals (CI)

Fig. 2 Forest plots of netting effect size (%) on total acidity (TA), starch index and colour (°hue angle) across cultivars in major fruit growing regions worldwide; error bars are 95% confidence intervals (CI) highly significant (p < 0.001) positive effect of 0.078 (95% CI 0.054–0.102).

The acidity of apples of the cultivar Fuji was, to a lesser extent, but *statistically highly significant* (p = 0.010) and negatively affected by netting -0.019 (95% CI -0.027 to -0.011) (Table 2). Pinova also had statistically highly significantly (p < 0.001) less acidic fruit under netting by -0.129 (95% CI -0.159 to -0.098), the third reason to exclude this cultivar from cultivation under netting.

#### **Starch conversion**

There was *no overall statistically significant* (p=0.659) tendency of netting on starch degradation. Differences between the studied cultivars, however, were statistically highly significant (p < 0.001). A statistically highly significant (p < 0.001) positive effect on starch conversion was observed in the two late ripening cvs Braeburn i.e. 0.108 (95% CI 0.095–0.121) and in Cripps Pink with an effect of 0.181 (95% CI 0.161–0.201), a very positive effect of earlier maturation in these late ripening cultivars, which is desired at the end of the season with the risk of an autumn frost and snow on hillsides of higher altitudes and/or latitudes.

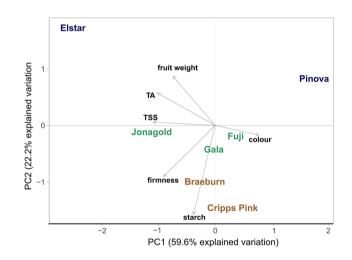
In Pinova, there was a *highly significant* (p < 0.001) *negative effect of netting on starch* degradation of -0.148 (95% CI -0.185 to -0.111) (Fig. 2), i.e. delaying ripeness and harvest, another negative effect of this cultivar, which seems outstanding and controversial to the others (Table 2).

#### Fruit colour

There was a statistically highly significant (p < 0.001) general positive effect of 0.077 (95% CI 0.043–0.111) of netting on hue angle, with a higher hue angle representing undesirable less red colour of the fruit peel. The difference of the effect on individual studied cultivars was also highly significant (p < 0.001). In cultivar Fuji, there was a highly significant (p < 0.001) effect size of 0.109 (95% CI 0.078–0.1403). Another highly significant (p < 0.001) effect size was observed in cultivar Gala 0.078 (95% CI 0.034 to 0.122) and cultivar Pinova 0.159 (95% CI 0.065–0.254) (Fig. 2), which suffered most from lack of red colouration; the use of reflective mulches (such as Extenday, Daybright or Lumilys) (Solomakhin and Blanke 2007) or easy red colouring mutants e.g. Gala Galaxy over Gala Mondial or Fubrax over Fuji Kiku 8, Rosy Glow over Pink Lady in Cripps Pink, could overcome this adverse effect.

#### Main effect meta principal component analysis

Main effects of netting in all studied cultivars with their characteristics recorded as metadata were analysed through Main Effect Meta-PCA (Fig. 3), explaining 82 percent of variations in the first two principal components. Along with the first Principal Component (60% of variation explained) (Fig. 3), the greatest influence or change was in total soluble solids (TSS) and in fruit colour, while the second component



**Fig. 3** Main effects meta-PCA on the effect of netting on studied fruit quality characteristics across studied cultivars (axes PC1 and PC2 represent 82% of data variation)

 Table 2
 Summary of results-netting effects on plant physiology and fruit quality

Location	P value	Overall effect size	Confidence interval (CI 95%)	Effect	Significance
Fruit firmness***	< 0.001	-0.019	-0.031 to $-0.009$	Small negative	Significant
Fruit weight*	0.0339	+0.023	+0.007 to $+0.041$	Small positive	Significant
Soluble solids (TSS)***	< 0.001	-0.038	-0.047 to $-0.029$	Small negative	Significant
Acidity (TA)	0.2220	-0.018	-0.049 to $+0.011$	Small negative	Not significant
Starch breakdown	0.6598	+0.007	-0.025 to $+0.039$	Small positive	Not significant
Colouration (°hue)***	< 0.001	+0.077	+0.043 to $+0.111$	Positive (i.e. less colour)	Significant
Delay or shorten ripening					
Early to medium varieties	Gala	Jonagold	Unaffected		
Late varieties	Braeburn	Cripps Pink	Earlier ripening		

Asterisks denote the following significance level: \*\*\*p<0.001; \*\*p<0.01; \*p<0.05

(22% of explained variation) was mostly influenced by starch index alterations due to netting cover. The grouping of cultivars Jonagold, Fuji and Gala (green in Fig. 3) can be observed in the middle of the graph, indicating general stability of those cultivars regarding the netting effect (Fig. 3).

The second group of cultivars consisting of the late ripening Braeburn and Cripps Pink is clustered along the second axis (brown in Fig. 3), indicating its general sensitivity regarding the starch content. Cultivars Pinova and Elstar are separated from the two main groups. Pinova (blue) is under influence of netting prone to loss of fruit firmness, delayed starch conversion, loss of soluble solids (sugar) and loss of colour, whereas Elstar (blue) may benefit from both more acidity and fruit weight in combination with decreased firmness (Fig. 3), particularly in years of alternate bearing (Krasniqi et al. 2013).

The effects on fruit characteristics are also grouped. Most characteristics reflected their impact due to netting effect along the negative part of the first principal component (left hand) around the TSS, which is positively correlated to acidity (TA), fruit weight and fruit firmness (Table 3). Fruit colour (or its loss as °hue) is the single characteristic defining the positive part of the first principal component and was negatively correlated with sugar TSS ("less colour less sweet") and the group of characteristic that predominantly defines the second principal component (right hand side in Fig. 3), had a borderline and significant positive correlation with fruit firmness (Table 3).

# Discussion

Netting alters the microclimate in the orchard (Fig. 4) and plants can acclimate to such altered environmental conditions, or adapt by different genetic response to such changes (Jackson 2003). The effect of hail netting on apple fruit characteristics was intensively researched, but the results of the published research were contradictory, which made it increasingly difficult to draw any general conclusions regarding the fruit characteristics in different cultivars. The simple at a first-glance solution is the biplot of the ME Meta-PCA (Fig. 3), which presents the overall effects of hail netting on



Fig. 4 A Fuji orchard at Klein-Altendorf nr Bonn under hailnet with such altered microclimate and ecosystem (© M. Blanke, Bonn)

apple fruit characteristics across a range of the most widely grown cultivars. With 82% of the explained variation in the presented graph, it provides a deep insight without much trade-offs.

Netting can reduce the fruit surface temperature by 1.5-3 °C due to shading, i.e. interception of solar irradiation and its scattering effect (Fig. 4) in combination with minimal airflow under the netting (McCaskill et al. 2016; Solomakhin and Blanke 2010). Leaf temperature is affected due to decreased transpiration and water flow through leaves under net (Solomakhin and Blanke 2010). Netting reportedly decreases photosynthesis (Amarante et al. 2009; Romo-Chacon et al. 2007; Solomakhin and Blanke 2008; Stampar et al. 2001), which reflects the deterioration in leaf structure (Asada and Ogasawara 1998) with fewer palisade layers (Solomakhin and Blanke 2010). The specific environment under netting in comparison with the open field microclimate (control) is characterized by generally increased humidity, lower temperature and decreased airflow (Iglesias and Alegre 2006; McCaskill et al. 2016; Middleton and McWaters 2002; Solomakhin and Blanke 2007). As temperature and solar irradiation are the most important climatic factors affecting fruit maturation (Ferree and Warrington 2003), netting impacts fruit maturation and related

Table 3Correlation coefficientsbetween the studied fruitcharacteristics under netting andprincipal components

	Colour	Firmness	Fruit weight	Starch	Acidity	TSS	PC1	PC2
Colour	1.000	-0.501	-0.241	-0.042	-0.567	-0.681	0.729	-0.089
Firmness	-0.501	1.000	0.257	0.641	0.615	0.813	-0.769	-0.517
Fruit weight	-0.241	0.257	1.000	0.004	0.779	0.637	-0.693	0.465
Starch	-0.042	0.641	0.004	1.000	0.051	0.294	-0.364	-0.867
Acidity (TA)	-0.567	0.615	0.779	0.051	1.000	0.947	-0.903	0.297
Sugar (TSS)	-0.681	0.813	0.637	0.294	0.947	1.000	-0.968	0.011

characteristics in two ways, i.e. light reduction and temperature reduction. However, the response differs between cultivars, as it has been reported regarding the difference between fruit and leaf surface temperatures in a comparative study on Fuji and Pinova, with inverse effects on leaf and fruit between the studied cultivars on sunny and cloudy days (Solomakhin and Blanke 2010). This study has also indicated that there are responses to hail netting, which are predominantly cultivar dependent.

## Impact on cultivars

The majority of alterations in fruit characteristics of studied cultivars influenced by netting were statistically significant. Although the majority of those changes was generally small rendering it of low practical importance, there were several distinguishing exceptions. Cultivar Gala proved to be the most stable with only a statistically significant substantial problem with fruit colouration and minor statistically significant alteration in soluble solids content. In the Fuji cultivar, there was also only one practically important issue of fruit colouration under netting, while there were no major changes in the other fruit characteristics of Fuji apples. Both Fuji and Gala fruit develop a specific deep colour, which in case of Fuji, requires a long maturation time to develop (Ferree and Warrington 2003). It is reported that under netting chlorophyll decomposition in the epidermis affecting fruit colour takes an average of 7 days longer in Gala and 10 days longer in Fuji (Brglez Sever et al. 2015; Germsek and Unuk 2014). The length of the season in terms of time each individual cultivar requires to reach fruit maturity, varies mostly in accordance to temperature fluctuations (Jackson 2003). As netting alters the temperature, it possibly affects the length of the season and consequently the harvesting date. Jonagold has no major fluctuations across fruit characteristics, except for colour. Cultivars Gala, Fuji and Jonagold were obviously grouped together in the centre of the final Fig. 3, hence characterized as generally stable with coloration issues.

In Braeburn, there is a statistically significant netting effect on starch and soluble solids content. Starch index is 10 percent higher due to netting, which is also of practical significance and a basis for consideration when planting this cultivar under hail or shade netting. Cripps Pink fruit also proved sensitive regarding the starch index and breakdown. Both Braeburn and Cripps Pink are cultivars with firmer crisp consistency and need more time to mature (Ferree and Warrington 2003; Jackson 2003). Those two long-season cultivars grouped together showed advanced starch breakdown under the netting.

Elstar apple is short-season apple distinguishable by its smaller fruit with higher sugar and acid content (Jackson 2003). Netting in Elstar led to significantly larger fruit, probably in the year of its alternate bearing and increased acidity. Pinova cultivar is also specific regarding netting effects: It is the most sensitive cultivar prone to significant changes due to netting for majority of measured fruit characteristics.

# Morphological response to netting and grouping of fruit characteristics

There are two main types of stress that have to be taken into consideration when comparing fruit characteristics of plants under netting to those in the open environment. First is the constant exposure of netted plants to decreased solar irradiation. Second type is the stress of occasional high temperatures and consequential higher temperature amplitudes that impacts plants in the opened environment, which is mitigated by shade in the plants under the nets. Both light and temperature are categorized among the most important factors affecting biochemical, physiological and morphometric aspects of fruit development (Flaishman et al. 2015; Li et al. 2015; Zhao and Guo 2011). Although there is limited knowledge on primary receptors that actually sense these stresses, signalling pathways are identified (Zeller et al. 2009) and as such can be used to explain consequential morphometric alterations. Reasoning resulting from this study on apple may be analogously transposed to other species grown under protective netting such as blueberry and kiwi, as fruit characteristics of interest and revealed mechanisms are often similar.

Responses of plants to mild chronic stress involves several mechanisms leading to morphologic changes which are all reportedly related to induction of calcium and reactive oxygen species (ROS) and their crosstalk to auxin and ethylene (Potters et al. 2009; Salopek-Sondi et al. 2017; Zeller et al. 2009; Zhao and Guo 2011). Those morphometric responses, however, seem to be interchangeable in the way that many different signals from the environment leading to different physiological and biochemical mechanisms lead to same metabolic and morphologic status (Potters et al. 2009) with even overlapping genes found to react on different types of stress (Zeller et al. 2009). Given that one of the most important environmental signals for plant growth and development is light (Zhao and Guo 2011), it becomes evident that shading in netted apples induces mild chronic stress by constantly reducing solar irradiation (Fig. 4). Decreased amounts of light even in form of fog, rain or mist adversely affect fruit size and also lead to decreased sugar content (Li et al. 2015), which is found in this study for apples under the hail netting.

The second type of stress is temperature; in this case, netting protects the plants or trees from excessive temperature stress (McCaskill et al. 2016; Solomakhin and Blanke 2010). The main effect of temperature stress is on reproductive tissues, i.e. fruit (Agarwal et al. 2017), as temperature alterations affect fruit growth—especially the cell division

rate, fruit ripening and its chemical composition (Saudreau et al. 2011). Similar to other stress response mechanisms temperature changes in plants after perceived by receptors are then transduced into cellular nucleus inducing changes in gene activation and transcription which ultimately leads to plant response in terms of physiological and biochemical alterations, often reflected as morphometric changes (Agarwal et al. 2017; Zeller et al. 2009). One of the main hormones involved in regulation of heat stress is ethylene, which is reportedly down-modulated in reproductive tissues as response to heat stress (Savada et al. 2017). It is well established that temperature is one of the key factors affecting fruit size, colour, sugar content, acid content and overall nutritional quality (Saudreau et al. 2011). Therefore, temperature alterations have to be considered in detail while discussing comparisons between netted and open orchards. Higher temperatures reportedly increase the speed of fruit development (Li et al. 2015), but may adversely affect fruit growth (Flaishman et al. 2015).

Sharp distinguishing between the morphological responses to the two main types of stress is not possible as physiological pathways are interchangeable and often lead to same morphometric response (Potters et al. 2009). It is reported that genes regulating heat response overlap to the largest extent with genes responsible for other types of stresses (Zeller et al. 2009). This however enables mitigation response that possibly covers several of those triggers by interfering with the undesired pathways and leading to the desired metabolic state.

# Conclusion

The meta-analysis of initially ca. 200 publications with 11,277 individual data recordings from 537 observations of concomitant comparisons of both netted trees and the control (adjacent trees without hail net) from the majority of fruit growing regions worldwide (from Germany (Bonn) to Tasmania, South Africa to Poland, Chile to Switzerland) provided surprisingly uniform results. Unexpectedly, climate, planting density and hail net colour (bright or dark) did not have major effects on the influence of the hail net on fruit quality parameters in comparison to the effect of individual cultivars. The outstanding results were (a) the correlation between red fruit colouration and sweetness, as often postulated in consumer studies (Hamadziripi et al. 2014), (b) identification of Gala and Jonagold as the least affected by netting and (c) the earlier fruit maturation under netting with the two late-season cultivars, Braeburn and Cripps Pink, where this is a valuable asset. This study also identified one cultivar (Pinova) very clearly and uniformly as unsuitable for cultivation under netting due to loss of fruit firmness, sugar, colour and delayed starch degradation and ripening; otherwise, these influences are all acceptable for apple cultivation under netting as a consequence of climate change to combat increasing hail storms and can be overcome by red mutants or reflectants.

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