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ORIGINAL PAPER

THE INFLUENCE OF A SOIL ACTIVATOR CONTAINING HUMIC ACIDS ON THE YIELD AND QUALITY OF APPLES IN CONDITIONS OF REPLANTATION

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Abstract

One of the ways of improving the physicochemical properties of replanted soil is to use fertilisers containing humic acids. The aim of the study was to analyse the impact of soil activator H-850 WG on the yield and quality of apples of the Najdared cultivar harvested in conditions of replantation. Between 2015 and 2017 an experiment was carried out on replanted and crop rotation soils. The replanted soil came from an orchard where apple trees had grown for several dozen years and they had been replanted several times. At the crop rotation site in the apple orchard there was a ten-year cultivation break, during which plants exhibiting the phytosanitary effect were grown (wheat, rape and charlock). A soil activator containing more than 60% of humic acids was used on both soil types. The following parameters were analysed in the experiment: the yield volume, fruit quality (average weight, extract content, fruit firmness and acidity level), the leaf surface and the content of chloroplast pigments in it. The results of the three-year research showed that the previous soil use method influenced the yield, the leaf assimilation area and the quality parameters of apples. The parameters analysed in the study were significantly worse on the replanted soil. The treatment of the soil with the activator containing humic acids resulted in a greater yield and better qualitative parameters of apples – increase the average fruit weight, extract content and fruit firmness. The soil activator exhibited higher efficiency on the replanted soil than on the crop rotation soil.

Keywords: replantation disease, humic acids, apple yield, extract content, fruit firmness and acidity.

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INTRODUCTION

The apple tree is the most important species for fruit production in Poland. In 2019, the national production of apples was estimated at about 2.8 million tonnes, which made Poland the world's fourth largest producer (following China – about 42 million tonnes, the USA – about 5 million tonnes and Turkey – about 3 million tonnes). There are not enough new, unused sites in regions of intensive fruit production in Poland. Due to consumers' changing preferences and the natural ageing of trees, apple growers need to plant new trees in the places where old trees have been felled. This increases the risk of soil fatigue, which leads to replantation disease. It is noteworthy that apple trees are particularly vulnerable to its effects. Replantation disease is manifested by a worse soil structure, higher soil acidity (Khan et al. 2008), poorer plant growth (Zydlik, Zydlik 2013, Sobiczewski et al. 2018), delayed fruiting, and lower yield from trees (Zydlik et. al. 2019). Replanted soil is also characterised by worse biological properties (Pacholak et al. 2009, Van Schoor et al. 2009). This is manifested by the imbalance of soil microorganisms. In consequence, there are excessive amounts of soil pathogens, e.g. fungi of the *Cylindrocarpon*, *Rhizoctonia*, and *Pythium* genera (Kelderer et al. 2012), *Pratylenchus penetrans* nematodes (Mazzola, Manici 2012), and phytotoxic phenolic compounds from the remains of tree roots (Politycka, Adamska 2003, Styla 2014).

It may take several years to restore the fertility of replanted soil. It is not always possible to clearly identify the cause of replantation disease due to the high diversity of causative factors. Therefore, it is also difficult to mitigate its effects. Apart from obvious methods such as not planting trees in the place where old trees grew before, the effects of replantation disease can be mitigated by improving the physicochemical and biological properties of soil. This can be done by means of organic fertilisers, which stimulate soil bioactivity by increasing the diversity of beneficial microorganisms and by reducing the amount of soil pathogens (Lazarovitz et al. 2001). However, this natural process of humification of organic matter applied into soil with manure is slow and laborious. Another option is to apply a mineral fertiliser, but it may reduce the population of soil bacteria (Zydlik 2010).The humus content in soil can be increased faster by means of soil activators, which contain mainly substances of natural origin, including humic acids. Apart from natural sources of humic acids, there are also commercially available humic preparations. They are usually made from leonardite, which is a lignite oxidation product.

Humic acids stimulate the development of the plant root system by increasing its length and the number of side roots (Conselvan at al. 2017). They improve the soil retention capacity, which ensures better conditions for plant growth and development (Selim, Mosa 2012). They also facilitate the uptake of soil nutrients (Naidu et al. 2013, Denre et al. 2014, Ennab 2016), improve plant growth and yield (Cangi et al. 2006, El-Bassiouny et al. 2015). These effects are achieved by increasing the productive value of soil. Humic acids increase the content of organic matter in soil and improve its biological properties, e.g. the amount of beneficial microorganisms (Rombel-Bryzek, Pisarek 2017) and enzymatic activity (Zydlik, Zydlik 2020).

The aim of the study was to analyse the effect of the H-850 WG soil activator containing humic acids on the yield and quality of apples of the Najdared cultivar harvested in conditions of replantation.

METHODS

Between 2015 and 2017, an experiment was conducted on apple trees of the Najdared cultivar grafted on the M26 rootstock. The research site was an orchard at the experimental station of the Poznań University of Life Sciences in the village Przybroda (52°31′ N, 16°38′ E). The orchard was established in 2009. The trees were spaced at 3.33 × 1.5 m (2.000 trees ha^{-1}) in two types of sites. The first site was so-called replanted soil with trees planted in the place where old apple trees had grown since 1949. During several decades of cultivation, trees were replanted several times. The last apple orchard was grubbed up in autumn 2008. In the following year, a new apple orchard was planted. The field with the orchard was not prepared in any way except except mineral nutrient supplementation. The occurrence of soil fatigue in this place was confirmed by previous results of the authors' research (Zydlik 2012). The other site included the soil in which apple trees had also grown since 1949, but they were not cultivated between 1998 and 2009 due to crop rotation. During that period, phytosanitary plants (wheat, rape and scharlock) were grown. *Endocalcaric Cambisol* (sandy loam soil) (WRB 2014) was the soil type at both sites. The content of nutrients in the soil and its biological properties were presented in an earlier publication of the authors (Zydlik, Zydlik 2020). The course of the weather conditions during the experiment is shown in Table 1.

The H-850 WG soil activator containing 65% humic acids, 10% fulvic acids and water-soluble K_2O was used in the experiment. The preparation was sprayed on the soil under the tree crowns three times during the growing season (in mid-April, early May and early September) as a one-time dose of 3 kg ha^{-1} for each treatment.

The following combinations were used in the experiment: 1 – replanted soil, 2 – replanted soil treated with H-850 WG, 3 – crop rotation soil, 4 – crop rotation soil treated with H-850 WG. All combinations in the experiment included two blocks with 20 trees in each. The following measurements and analyses were made during the three years of the research: leaf area, chlorophyll and carotenoid content in leaves, percentage of fruit set, fruit yield (kg per tree) and quality (weight, firmness, extract content, acidity). All analyses were conducted in four replications.

Table 1

The course of climatic conditions in the years 2015-2017

Years	Months						Sum
	April	May	June	July	August	Sept.	
Precipitation (mm)							
2014	41.8	65.2	33.0	48.4	93.6	36.6	318.6
2015	18.0	28.8	58.6	78.6	15.2	31.2	230.4
2016	35.4	16.4	121.2	120.8	37.4	18.6	349.8
2017	36,4	31,2	85,6	182,4	80,0	47,2	462,8
1978-2007	30.3	45.5	64.5	77.9	59.6	45.2	323.0
Air temperature (°C)							mean
2014	10.6	13.3	16.1	21.6	17.4	15.2	15.7
2015	8.5	12.9	15.7	19.1	22.0	14.2	15.4
2016	8.9	15.2	18.2	18.9	17.5	16.3	15.8
2017	7,8	13,3	17,5	17,8	18,2	14,1	14,83
1978-2007	8.6	14.3	17.1	19.1	18.6	13.9	15.3

50 inflorescences were counted on selected, marked shoots on the trees in each treatment in order to calculate the percentage of fruit set. During the harvest, the number of fruit harvested from these shoots was counted. The harvest date was determined according to the degree of starch degradation.

The qualitative parameters were evaluated on 50 randomly selected fruits from each treatment. The fruits were weighed with an accuracy of 1 g. The firmness (kg cm^{-2}) was measured using a Fruit Pressure Tester mod. 327 firmness meter (Faccini). A pin with a diameter of 11 mm was thrust into the apple flesh to a depth of 8 mm at a site of intense ruddiness and at a site of the basic skin colour. The extract content in the same fruits was determined with a refractometer. In order to determine acidity (the percentage of malic acid), 45 ml of distilled water was poured over 5 ml of juice and then titrated with 0.1 N NaOH to pH 7.4.

Leaves were collected from trees only once during the growing season, in the second half of July. 10 leaves were harvested from the middle part of annual long shoots of each tree in a treatment (200 leaves per treatment). The area of the leaf (cm^2) was scanned and measured with the DigiShape program. The content of chlorophyll *a* and *b* in the fresh mass of leaves was measured by extraction with dimethyl sulphoxide. A weighed amount of 0.5 g of fresh leaves was taken for tests. 8-mm discs were cut out with a cork borer. Next, 5 ml of dimethyl sulphoxide was poured on them. Before measurements, the samples were placed in a water bath at 65°C for 20 min so that chlorophyll would go into the solution. Next, the solution was analysed spectrophotometrically at a wavelength of 470 nm for carotene, 645 nm for chlorophyll *b* and 663 nm for chlorophyll *a*. The results of the analyses were subjected to two-way analysis of variance and Duncan's test at a significance level $\alpha = 0.05$. The results of the yield were analysed using profile analysis and canonical variate analysis (Kayzer et al. 2015). The canonical variate analysis method consists in the transformation of the original set of variables into a set of new variables, which carry similar information, but are distributed in a multivariate Euclidean space (Lejeune, Calinński 2000, Kayzer et al. 2009). In this case, this method is based on the matrix including the differences between mean values of annual yield for the considered stand types and the general annual means.

RESULTS

The experiment showed that the yield of the Najdared apple trees was affected by the previous method of soil use. During the three years of the research, the average yield of the trees growing on the crop rotation soil was almost three times greater (15.39 kg per tree) than the yield of the trees growing on the replanted soil (4.36 kg per tree) – Table 2. The relationship

Table 2

The influence of the experimental factors on the yield of apples (kg per tree) in the years 2015-2017

Type of soil	Humuc acids	2015	2016	2017	Average
Replanted	-	5.40*b**	2.488*a*	5.21*b*	4.36*A***
	+	13.58*d*	6.30i	10.78*c*	10.22*B*
Crop rotation	-	10.88*c*	14.45*d*	20.84*f*	15.39*C*
	+	16.00*de*	17.7*e*	24.35*g*	19.35*D*

* means marked with the same small letter do not differ significantly at $\alpha = 0.05$
** means marked with the same capital letter in columns do not differ significantly at $\alpha = 0.05$

between the treatments used in the experiment and the yield of the apple trees is shown in Figure 1. The analysis of the diagram also shows clearly that the yield of the apple trees growing on the crop rotation soil was significantly higher than the yield of the trees growing on the replanted soil. A decrease in the yield of apple trees growing on replanted soil was also observed by Pacholak et al. (2009) and Zydlik (2012).

There were higher yields of the apple trees in the treatments with the soil activator, especially those growing on the replanted soil. During the three years of the research, the average yield of the trees growing at that site was twice as high (10.22 kg per tree) as in the treatments where the preparation had not been used (4.36 kg per tree) – Table 2. The differences in the yields of the trees growing on the crop rotation soil treated and untreated with the soil activator were smaller, less than 25%. The findings

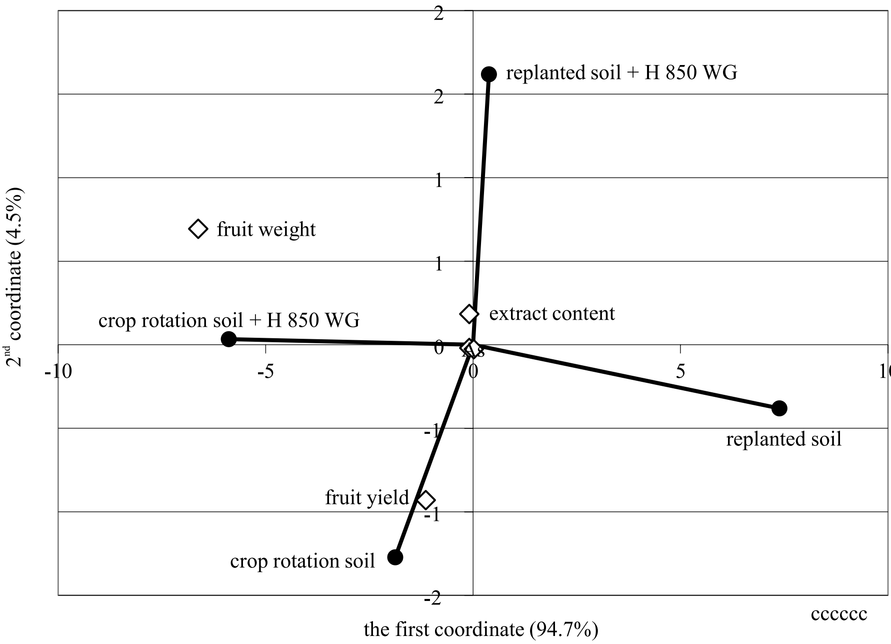

Fig. 1. The yield and qualitative characteristics of apples vs the combinations used in the experiment

of our study are similar to the results of the research conducted by other authors, who observed that humic acids increased the yield of pears (MANSOUR et al. 2013), apricots (FATHY et al. 2010) and strawberries (FARAHI et al. 2013).

The differences in the yield of the Najdared cultivar apple trees may have also been influenced by other factors analysed in the experiment. Between 2015 and 2017, the average percentage of fruit set on the trees growing on the replanted soil was four times lower (12%) than on the trees growing on the crop rotation soil (51.25%) – Table 3. The soil activator used in the experiment significantly modified this parameter. The average percentage of fruit set on the trees growing on the replanted soil increased from 12% to 36%, whereas on the crop rotation soil it increased from 51.25% to 67.83% (Table 3).

Another factor that influenced the yield of apple trees was the leaf assimilation area and photosynthetic activity, measured with the content

Table 3

The influence of the experimental factors on the percentage of fruit set in the years 2015-2017

Type of soil	Humic acids	2015	2016	2017	Average
Replanted	-	11.25	13.53	11.25	12.00
	+	19.25	48.00	40.75	36.00
Crop rotation	-	41.50	54.52	57.75	51.25
	+	50.25	77.75	75.50	67.83

of chloroplast pigments. These parameters also varied significantly depending on the site. Between 2015 and 2017, the average leaf area on the Najdared apple trees growing on the crop rotation soil (25.48 cm^2) was almost twice as high as that of the trees growing on the replanted soil (14.18 cm^2) – Table 4. Sobiczewski et al. (2018) also observed a small leaf assimilation area in trees growing on replanted soil. The treatment of the soil with the activator increased the leaf assimilation area of the apple trees. During the three years of the research, the average leaf assimilation area of the trees growing on the replanted soil treated with the H-850 WG preparation was over 100% higher than in the combination without this treatment (14.18 and 30.81 cm^2) – Table 4. By comparison, this increase did not exceed 30%

Table 4

The influence of the experimental factors on the leaf area, chlorophyll and carotenoid content in leaves in the years 2015-2017

Type of soil	Humic acids	2015	2016	2017	Average
Leaf area (cm^2)					
Replanted	-	9.06*a**	15.23*b*	18.25*bc*	14.18*A***
	+	21.79*c*	33.81*d-f*	36.85*f*	30.81*C*
Crop rotation	-	16.41*bc*	30.08*de*	29.95*de*	25.48*B*
	+	28.03*d*	35.55*ef*	38.98*f*	34.18*D*
Chlorophyll *a* + *b* (mg kg^{-1} fresh mass)					
Replanted	-	157.85*c*	157.85*c*	156.53*b*	157.41*A*
	+	158.90*c-e*	158.01*c*	154.98*a*	157.29*A*
Crop rotation	-	159.13*de*	158.40*cd*	155.00*a*	157.51*A*
	+	159.50*e*	158.14*cd*	154.41*a*	157.35*A*
Carotenoids (mg kg^{-1} fresh mass)					
Replanted	-	564.93*a*	760.69*c*	761.30*c*	695.64*A*
	+	611.73*ab*	708.66*bc*	811.68*cd*	710.69*AB*
Crop rotation na	-	777.32*cd*	708.96*bc*	810.82*cd*	765.70*BC*
	+	884.78*d*	709.76*bc*	810.45*cd*	801.66*C*

*, ** remarks as to Table 2

in the trees growing on the crop rotation soil (from 25.48 to 34.18 cm^2). A similar positive effect of humic acids on the growth of leaf surface was recorded by Ennab (2016).

The proper course of photosynthesis in leaves depends on their chlorophyll content. This process is decisive to the accumulation of biomass in plants. Available publications do not provide much information about the influence of replantation disease on photosynthesis in apple leaves. In our experiment the content of photochromatic pigments in the leaves of apple-trees did not vary significantly throughout the entire research period. The three-year average total content of chlorophyll *a* and *b* in leaves did not differ significantly regardless of the previous soil use method (Table 4). Only the content of carotenoids (the pigments that protect chlorophyll) in the leaves of the trees growing on the post-crop-rotation soil was slightly higher (765.70 mg kg^{-1} fresh weight) than the content in the leaves of the trees growing on the replanted soil (695.64 mg kg^{-1} fresh weight). Unlike the results published by Abourayya et al. (2020), the treatment of both types of soil with the preparation containing humic acids did not significantly change the content of chlorophyll ($a + b$) and carotenoids in apple leaves (Table 4, Figure 2). The results of our study differed from the findings of the research conducted by other authors, who observed that humic acids intensified photosynthesis (Cangi et al. 2006).

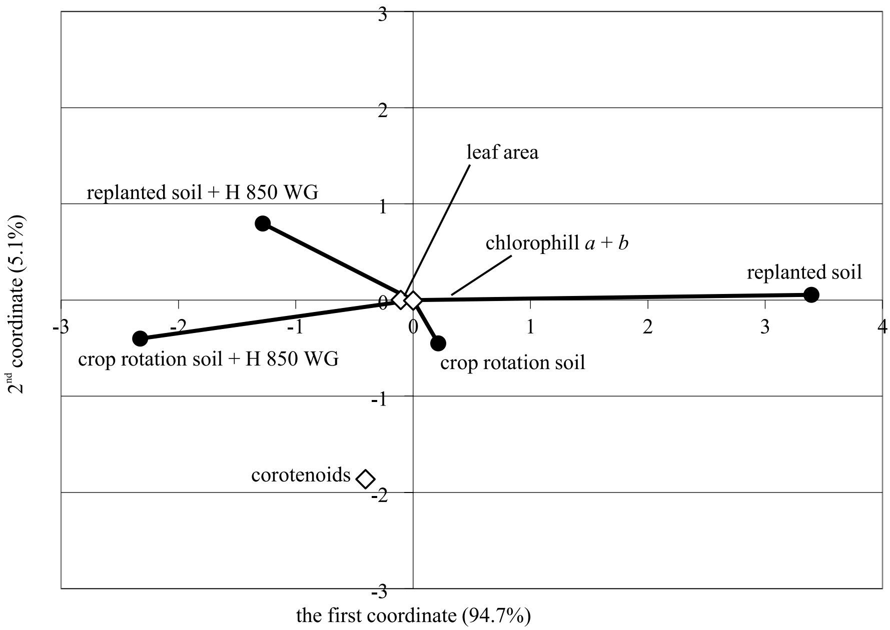

Fig. 2. The leaf area, chlorophyll and carotenoid content in leaves vs the combinations used in the experiment

The treatment of the soil with the activator containing humic acids could have resulted in a better supply of nutrients to the apple trees and increased their yield potential and leaf assimilation area. The H-850 WG preparation increased the content of $N\text{-}NO_3$, P, K, Mg and Ca both in the trees growing on the replanted soil and those growing on the post-crop-rotation soil, which was previously confirmed by the authors (Zydlik, Zydlik 2020). According to Ennab (2016), humic acids not only increase the content of organic matter in the soil, but also stimulate the proliferation of beneficial microorganisms and thus facilitate soil mineralization. The nutrients produced in this way can be easily absorbed by plants (Amery, Tehranifar 2012, Selim, Mosa 2012, Naidu et al. 2013, Denre et al. 2014, Zydlik et al. 2019). They are transformed into available forms, which can be easily absorbed by the cell membranes in the roots (Akinci et al. 2009, Nazli et. al. 2014, Conselvan et al. 2017).

The profitability and competitiveness of fruit production are determined by fruit quality. The quality of fruit is usually defined by the following parameters: average weight, firmness, extract content and percentage of acids. Our study showed that the previous soil use method influenced not only the yield of the trees but also the quality of their fruit. For example, between 2015 and 2017, the average weight of the apples from the trees growing on the crop rotation soil (214.3 g) was significantly higher than that of the trees growing on the replanted soil (184.42 g) – Table 5, Figure 1.

Table 5

The influence of the experimental factors on the fruit quality in the years 2015-2017

Type of soil	Humic acids	2015	2016	2017	Average
Average mass of fruit (g)					
Replanted	-	159.28*bc**	137.00*a*	149.00*ab*	148.42*A***
	+	174.93*cd*	225.00*g*	202.25*ef*	200.73*B*
Crop rotation	-	189.10*de*	235.43*gh*	220.25*fg*	214.93*C*
	+	202.95*ef*	253.00*h*	249.25*h*	235.07*D*
Firmness (kg cm^2)					
Replanted	-	6.25*bc*	5.20*a*	5.84*b*	5.76*A*
	+	6.50*cd*	6.22*bc*	6.63*cd*	6.45*B*
Crop rotation	-	6.60*cd*	6.63*cd*	7.00*d*	6.74*C*
	+	7.03*d*	6.93*d*	7.03*d*	6.99*C*
Soluble solid content (°Brix)					
Replanted	-	12.90*a*	14.60*cd*	13.28*ab*	13.59*A*
	+	13.70*b*	15.33*e*	15.10*de*	14.71*BC*
Crop rotation	-	13.83*b*	14.85*c-e*	14.67*cd*	14.45*B*
	+	14.43*c*	15.23*de*	15.10*de*	14.92*C*
	Acidity as the malic content (%)				
Replanted	-	1.39*f*	1.63*g*	1.28*e*	1.43*A*
	+	0.92*ab*	0.93*ab*	1.09*c*	0.98*A*
Crop rotation	-	0.96*b*	1.13*cd*	1.20*d*	1.09*B*
	+	0.88*a*	1.19*d*	1.14*cd*	1.07*B*

*, ** remarks as to Table 2

The fruits harvested from the trees growing at that site were characterised by significantly higher firmness (6.74 kg cm^{-2}) and extract content (14.45 °Brix) than the apples harvested from the trees growing on the replanted soil (5.76 kg cm^{-2} and 13.59 °Brix, respectively). Our results were different from the observations made by Zydlik (2010), who found that replantation disease did not significantly influence the quality of apples, measured with their firmness and extract content.

Our experiment showed that the soil activator containing humic acids had a positive effect on the qualitative parameters of apples, especially those harvested from the trees growing on the replanted soil. The average weight of the fruit harvested from the trees growing at that site was much higher than the weight of the fruit in the combination without this treatment (200.37 and 148.42 g or 235.07 and 214.93 g respectively) – Table 5. The average weight of the fruit harvested from the trees growing on the crop rotation soil was much higher than the weight of the fruit in the combination without the soil activator. The fruit firmness and the extract content also increased significantly. The positive effect of humic acids on the quality parameters of fruit was also recorded by BORAY et al. (2015). The percentage of acids in fruit were not influenced by different soil use methods (Table 5, Figure 1).

The relationship between the fruit yield and weight and the content of selected macronutrients in the soil is shown in Figure 3. In our study, Ca and Mg were the elements with the greatest influence on the weight of the fruits, especially those harvested from the trees growing on the post-crop-rotation soil). The effect of N-NO_3 on the fruit weight was much smaller, whereas the effect of P and K was unnoticeable (Figure 3).

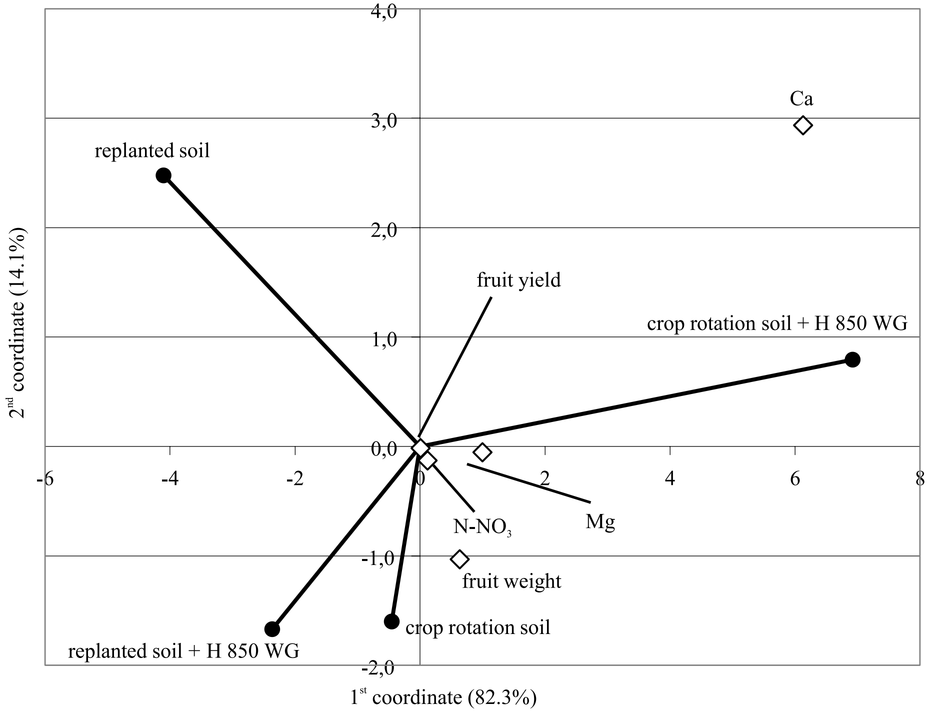

Fig. 3. The dependence between the soil nitrogen, calcium and magnesium content and the yield of apples

The preparation containing humic acids caused changes in the qualitative parameters under analysis. During the research, the average firmness and the content of extract in the apples harvested from the trees growing on the replanted soil increased by several per cent (from 5.76 to 6.45 kg cm^{-2} or from 13.59 to 14.71 °Brix respectively). The acidity of the apples decreased from 1.43% to 0.98% (Table 5). The high content of extract and low acidity of fruits determine their high value for desserts. Selim, Mosa (2012) or Denre et al. (2014) conducted an experiment on strawberries and observed an increase in their firmness under the influence of humic acids. The use of the activator on the crop rotation soil modified the qualitative characteristics of the fruit to a lesser extent. Neither the firmness nor the acidity of the fruit harvested from the trees growing at those sites changed significantly after treatment with the preparation (Table 5).

The size of fruit may affect its firmness and content of extract. This dependency was the subject of the research conducted by De Salvador et al. (2006). In our experiment, there was no significant correlation between the average weight of the fruits and their firmness and extract content. There was only an insignificant negative correlation between the average weight of the apples and their acidity (r = - 0.023, n = 47) (unpublished data).

Plants may differ in their response to humic acids. Not only is the species important, but so are the development phase, the method of supplying humic acids and the habitat conditions. Another factor analysed in our experiment was the course of climatic conditions. The weather significantly influenced the yield and quality of the apples. In the summer months (June-August) of 2017, the amount of rainfall was much higher than the long-term average (Table 1). In the same year, the highest yield of the trees was noted (Table 2). With such good humidity, the soil activator did not influence the yield of apples as much as in the previous two years (Table 2). In 2014, before the experiment, there was relatively low rainfall (Table 1). This resulted in relatively lower yields from the trees (Table 2) and a lower average fruit weight in the first year of the experiment (Table 5). The variability in the yield of the Najdared apple trees in different periods of the research cycle is shown in Figure 4. As seen, both the habitat and weather conditions influenced the yield. The first canonical coordinates represent 50% of the yield variation and are related to different climatic conditions in different years of the research. The second coordinates represent 44% of the variability and are related to the combinations used in the experiment. The analysis of canonical variability shows that it considerably influenced the average fruit weight. The location of the points determining the similarity of the combinations in individual years of the research in terms of the qualitative traits of the apples was determined by the course of the weather conditions (Figure 4).

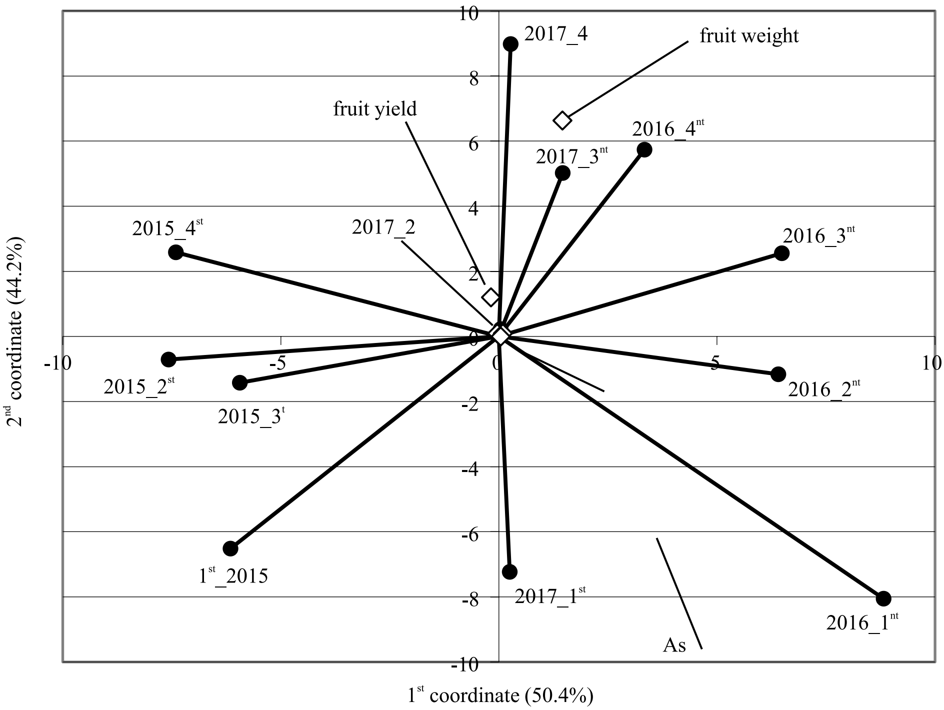

Fig. 4. The qualitative characteristics of apples vs the combination used in individual years of the research

CONCLUSIONS

1. The previous soil use method significantly influenced the yield of the apple trees. The yield of the trees growing on the crop rotation soil was three times higher than the yield of the trees growing on the replanted soil.

2. The apple trees growing on the replanted soil were characterised by significantly lower leaf assimilation area and fruit weight, lower fruit extract content and firmness, and lower percentage of fruit set than the trees growing on the crop rotation soil. There were no differences in the content of chloroplast pigments in the leaves.

3. The soil activator containing humic acids significantly increased the percentage of fruit set, the leaf assimilation area and the yield of the apple trees.

4. The soil activator positively influenced the qualitative parameters of the apple trees. It increased the average fruit weight, their firmness and extract content. The soil activator did not significantly affect the content of chlorophyll *a* and *b* in the leaves of the apple trees.

5. The soil activator increased the yield and improved the quality of apples more effectively on the replanted soil than on the crop rotation soil.

6. The weather conditions significantly influenced the yield of apples and their quality.

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