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Impact of Herbicides on Soil Biology in Rapeseed

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***Abstract*—**Winter oilseed rape, *Brassica napus* L., is characterized by a high number of herbicide applications. Therefore, its cultivation can lead to massive contamination of ground water and soil by herbicide and their metabolites. A multi-side long-term field experiment (EFFO, Efficient crop rotation) was set-up in Luxembourg to quantify these effects. Based on soil sampling and laboratory analysis, preliminary results showed reduced dehydrogenase activities of several soil organisms due to herbicide treatments. This effect is highly depending on the soil type. Relation between the dehydrogenase activity and the amount of microbial carbon showed higher variability on the test side with loamy Brown Earth, based on Bunter than on those with sandy-loamy Brown Earth, based on calciferous Sandstone.

***Keywords*—**Cropping system, dehydrogenase activity, herbicides, mechanical weed control, oilseed rape.

I. Introduction

WINTER oilseed rape, *Brassica napus* L. (WOSR), is an important element in crop rotation in the Grand Duchy of Luxembourg [1]. On average, 5,000 ha are grown per year in the country. The cultivating of WOSR is characterized by a number of advantages, such as:

- reliable yields of approx. 42 dt ha^{-1} [2],
- importance in grain based crop rotations [3],
- food resource for pollinator insects [4], and
- continuous soil cover throughout the year to prevent soil erosion [5].

Otherwise, the production of WOSR requires in most of the cases an intense usage of fertilizers and herbicides such as Metazachlor, Clomazone, and Pethoxamid [6]. Herbicides are used mainly when the WOSR plants are emerging and due to their young stages not very competitive to weeds. It was shown by regular ground and surface water-analysis that a high level of contamination by metabolites products of WOSR specific herbicides was found in Luxembourg, recently [7], [8]. Therefore, scientific, administrational, and advisory institutions of the agriculture business defined a multi-site multi annual cooperation project. The objectives of this project called EFFO ("efficient crop rotation") aims atthe identification of suitable cropping techniques to reduce the amount of highly mobile herbicides and transformation products used in WOSR.

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II. Material and Methods

Two experimental sites in Luxembourg were chosen based on their soil properties: Hobscheid (sandy-loamy calcic cambisol based on sandstone) and Reisdorf (loamy cambisol based on Bunter) (Fig. 1). Both soil types are typical - mostly fractured - aquifers in Luxembourg. On each of the sites, four different cropping techniques/treatments for WOSR were used in a randomised block design with four repetitions each (30 m^2 per parcel). The following cropping techniques/ treatments were implemented:

- V1: conventional WOSR with application of Metazachlor,
- V2: conventional WOSR with application of Clomazone plus Pethoxamid,
- V3: WOSR in biological cultivation in enlarged row space without any chemical control measures, and
- V4: WOSR with mechanical weed control (harrow).

WOSR (hybrid cultivar 'Exception') was sown in 31. August 2015 in a sowing density of 45 corn/m^2 after ploughing.

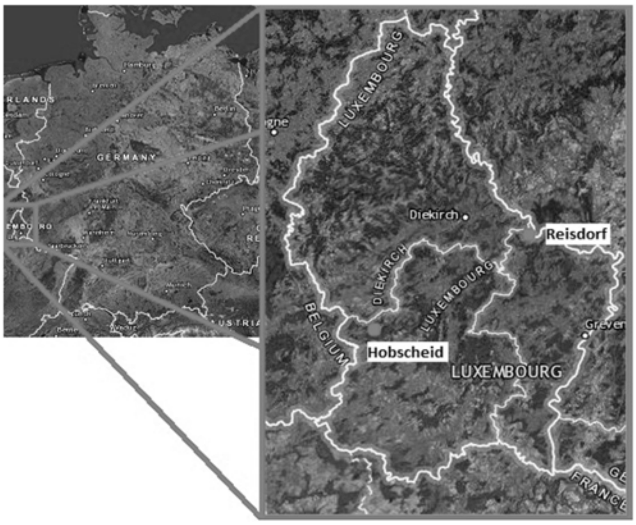

Fig. 1 Map of test side location in Luxembourg

Application of herbicides was done in two treatments before crop emergence on 4 September as follows: V1: Metazachlor (Butisan Gold, 2.5 l ha^{-1}), V2: Clomazone (Centium, 0.2 l ha^{-1}) plus Pethoxamid (Koban, 2 l ha^{-1}). In the biological treatment no chemical herbicides were used, but row space was enlarged up to 65 cm and weed control was done by using a rotary hoe within the rows, eight weeks after sowing (V3). A harrow destroyed weeds in the variety with mechanical weed control, when WOSR plants were in the growth stage of eight true leaves, six weeks after sowing (V4). Both experimental sites were equipped with standard meteorological weather stations with sensors for air temperature (2 m), relative humidity (2 m) and precipitation (1 m). All data were stored as minute values and an automatic data processing chain for quality control, gap detection and gap filling were applied. The quality control showed no gaps in the time series of the meteorological data for the period under investigation. For the later analysis, data were aggregated to daily values. Soil samples were taken on both locations two times, in autumn (October 2015) and in following spring (March 2016) in depth 0-30 cm with a soil probe (4 times per treatment, pooled sample). Samples were frozen by -18 °C. For measuring the soil water content of all samples, 10 g soil per sample was dried by 105 °C for 18 hours. Before and after the drying process, the weight of the samples was determined and the water content derived. The pH values were determined by using $CaCl_2$ as solvent.

For analysis the samples had been unfrozen and sieved (2 mm). Dehydrogenase activity was measured based on an already established method [9]. Results of Dehydrogenase are given as µg triphenylformazan (TPF) per gram dry mater. In the following analysis, the relation between dehydrogenase activity and the amount of microbial carbon (C_{mic}) was detected by the chloroform fumigation-extraction method [10].

III. Results and Discussion

The meteorological conditions during the field experiment at the different test sites were given in Figs. 2 and 3.

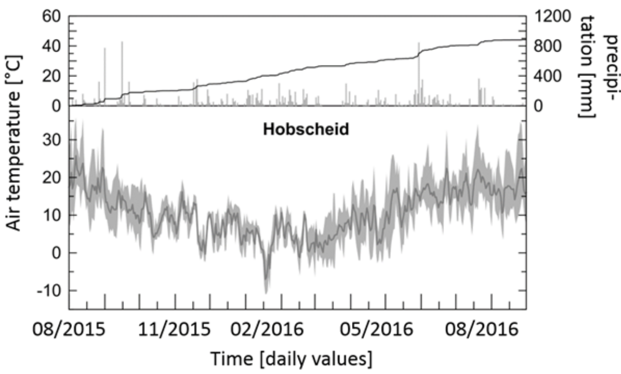

Fig. 2 Daily air temperatures and daily total of precipitation at the Hobscheid test site. Grey: spread defined via daily minimum and maximum air temperature. Black line upper part: cumulative precipitation sum

As given in Figs. 2 and 3, similar meteorological conditions were observed on both experimental sites, Reisdorf and Hobscheid. The mean air temperature – measured at a height of 2 m – at Hobscheid was 10.7 °C and in comparison, at Reisdorf 10.5 °C. Especially, the measurements at Hobscheid in Spring 2016 showed a higher daily variability in minimum and maximum air temperature values. The cumulative precipitation sums for Reisdorf amounts to 865 mm with very low precipitation sums in January 2016, whereas cumulative precipitation at Hobscheid was with 882 mm slightly higher.

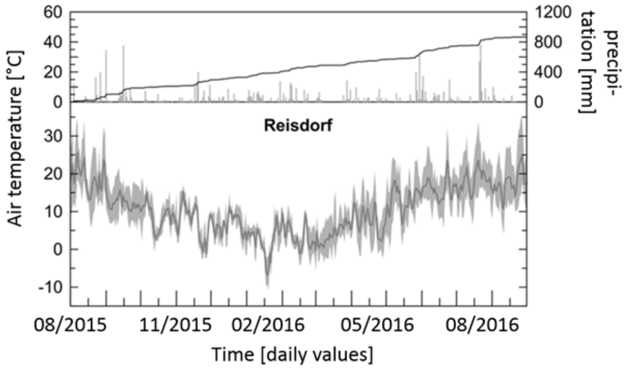

Fig. 3 Daily air temperatures and daily total of precipitation at the Reisdorf test site. Grey: spread defined via daily minimum and maximum air temperature. Black line upper part: cumulative precipitation sum

TABLE I
Soil Water Content (%) for the Two Test Sites, Hobscheid and Reisdorf in Autumn 2015 and Spring 2016 for Different Treatments and/or Cropping Techniques

treatment/cropping technique	autumn 2015	spring 2016	difference
Hobscheid			
V1 (Metazachlor)	21.2	16.3	4.9
V2 (Clomazone)	19.2	14.1	5.1
V3 (biological)	22.1	12.5	9.6
V4 (mach. Weed control)	21.3	15.0	6.3
Reisdorf			
V1 (Metazachlor)	29.5	22.3	7.2
V2 (Clomazone)	28.4	23.4	5.0
V3 (biological)	27.1	24.0	3.1
V4 (mach. Weed control)	27.9	26.8	1.1

TABLE II
pH Values for the Two Test Sites, Hobscheid and Reisdorf in Autumn 2015 and Spring 2016 for Different Treatments and/or Cropping Techniques

treatment/cropping technique	autumn 2015	spring 2016	difference
Hobscheid			
V1 (Metazachlor)	5.3	5.1	0.2
V2 (Clomazone)	5.2	5.0	0.2
V3 (biological)	5.8	5.3	0.5
V4 (mach. Weed control)	5.6	5.4	0.2
Reisdorf			
V1 (Metazachlor)	5.8	5.5	0.3
V2 (Clomazone)	5.6	5.3	0.3
V3 (biological)	5.9	5.8	0.1
V4 (mach. Weed control)	5.7	5.8	-0.1

The soils characteristics for the different test sites were described by soil water content (Table I) and the pH values (potential of hydrogen; Table II). The soil water content at Hobscheid showed typical values for a water enriched soil. The differences between the treatments are not significant. The sample from the biological cultivated WOSR showed the highest water content (22.1%), whereas the soil water content in the Clomazone treated WOSR was the lowest (19.2%). In comparison to autumn 2015, all samples had lower soils water content in spring 2016. At location Reisdorf for all soil samples, a water content between 27.1% and 29.5% was detected. Lowest values were measured in the soils samples taken from the biological cultivated WOSR (27.1%). As already detected at Hobscheid, the soil water content at Reisdorf was decreasing from autumn to spring in all treatments. Statistical significant differences were not detected. Comparing results from different experimental sites Reisdorf showed a higher soil water content (5%).

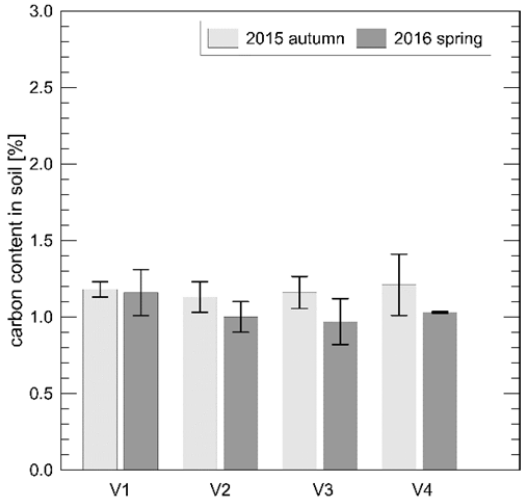

Fig. 4 Carbon content in percent of the soil from the different cropping techniques/treatment for autumn 2015 and spring 2016 at location Hobscheid

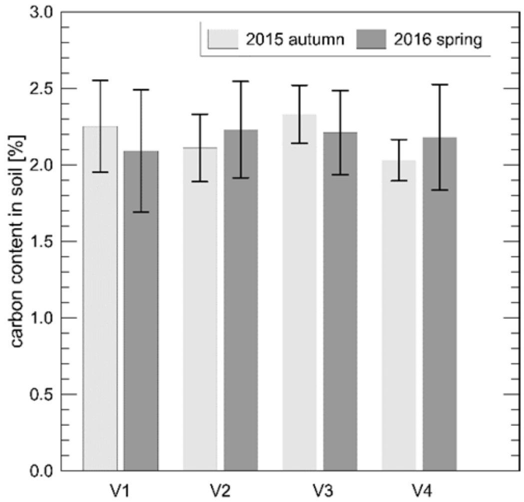

Fig. 5 Carbon content in percent of the soil from the different cropping techniques/treatment for autumn 2015 and spring 2016 at location Reisdorf

The pH value at location Hobscheid varied between 5.2 and 5.8 for the different treatments. The lowest pH-values was detected in the soil samples taken from the Clomazone treated WOSR and the highest in the mechanical weed control. No statistical significant differences could be detected in comparison to the location Hobscheid. At the location Reisdorf, the pH-values show only minor differences between the different treatments.

A comparison of the carbon content between autumn 2015 and spring 2016 is given in Fig. 4 for Hobscheid and Fig. 5 for Reisdorf. In comparison with the location Reisdorf, the carbon content for every cropping technique/treatment was reduced at location Hobscheid.

In Figs. 6 and 7, the results of the dehydrogenase activity for the different experimental treatments are given. At location Hobscheid, the dehydrogenase activity ranges from 53 µg TPF up to 61 µg TPF per g dry matter in 2015, except of the Clomazone treatment (V2). Due to the high variance of the samples, the results are not statistically significant. In spring 2016, the dehydrogenase activity showed no significant changes compared to the previous year. In the experimental plot treated with Metazachlor (V1), a slight increase was detected, whereas in all other test plots the dehydrogenase activity decreased compared to the autumn results (not statistically significant).

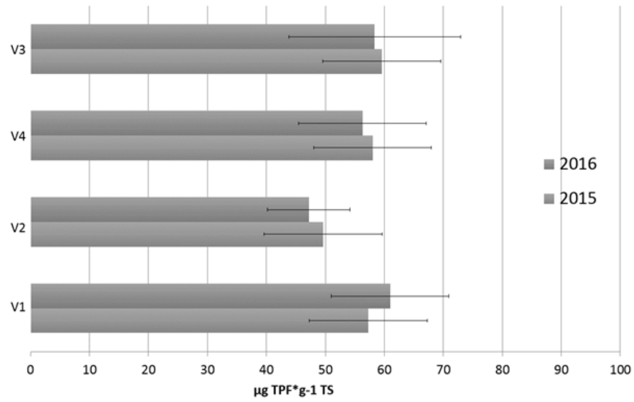

Fig. 6 Dehydrogenase activity for the different varieties given as µg Triphenylformazan (TPF) per g dry matter (TS) for the test side at Reisdorf in the 2015/2016 season

At the location Hobscheid, the test plot V4 showed the highest rates of TPF compared to the other plots. The differences between V2 and V4 are statistically significant. V1 and V2 (both treated with herbicides) showed lower enzymatic activity in comparison to V3 and V4.

Among all enzymes in the soil environment, dehydrogenases are one of the most important and are used as an indicator of overall soil microbial activity [11]. Because of the clay-humus-complex in the soil at location Reisdorf, herbicide molecules were absorbed at the soil colloids and therefore protected against chemical and physical degradation [12].

In comparison with the sandy soil at location Hobscheid, the bulk density of the clay soil at Reisdorf hampered an optimal oxygen level for microorganism; therefore, a lower dehydrogenase activity was detected. In the sandy soils at location Hobscheid, herbicide molecules are weaker for degradation because of absence of clay-humus-complexes

[13].

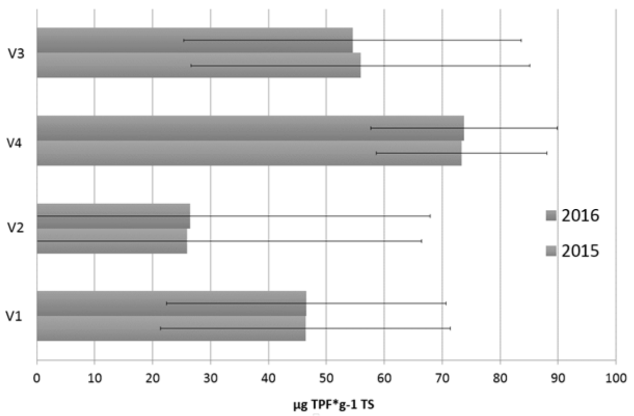

Fig. 7 Dehydrogenase activity for the different varieties given as µg Triphenylformazan (TPF) per g dry matter (TS) for the test side at Hobscheid in the 2015/2016 season

Using herbicides on sandy soils can have a significant effect on dehydrogenase activity [13], which is in line with our results: both herbicide treatments showed lowest levels of TPFs at location Hobscheid.

The effect of herbicides to enzymatic activity of soil organisms - demonstrated at both test sides - was showed in literature, recently [14].

The scatterplot in Fig. 8 shows the relation between the dehydrogenase activity and the amount of microbial carbon at both test sides. While at Hobscheid a high variability of the dehydrogenase activity and low variance in the microbial carbon was detected, at Reisdorf lower variabilities of both variables were measured.

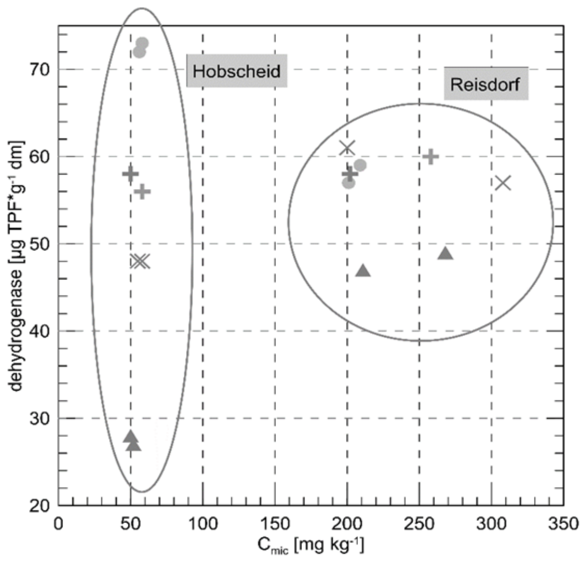

Fig. 8 Scatterplot of dehydrogenase activity and amount of microbial carbon (× = V1, ▲ = V2, ● = V3, + = V4)

These findings are in line with comparable studies [14], [15]. A more sustainable use of herbicides is necessary to ensure a high level of microbiological activity depending on the different soil types on Western Europe.

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