Effect of annual use of pesticides on soil microorganisms and sugar beet yields

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Abstract. Sugar beet is often cultivated for several years on the same fields, using many pesticides. We have therefore studied the effects of a pesticide programme on soil microorganisms and sugar beet yields in Perniö and Laukaa. The pesticides in use were thiram, hymexazol, dimethoate, phenmedipham and metamitron and, in Laukaa only, alloxidim-Na. Pesticides were used either in the normal doses or at 150\% of the normal dose.

The normal doses of pesticide application had a favourable effect on sugar beet yields in both experiments. The sugar yield was higher in plots with the normal pesticide doses than in the control plots. The overdoses increased neither the sugar beet yields nor the sugar yields as compared to the normal plots. The soil microorganisms were affected by pesticides in some but not in all cases. The most sensitive were the ureolytic microorganisms and the dehydrogenase activities.

Index words: Alloxidim-Na, dimethoate, hymexazol, metamitron, phenmedipham, thiram, total number, spore-forming aerobes, ureolytics, dehydrogenase, nitrification, cellulolysis

Introduction

The cultivation of sugar beet nowadays requires the use of pesticides. Sugar beet also requires a very good soil structure so that the developing roots get water and nutrients but do not lack air. For a long time in spring and early summer the soil is covered by crops only partly, and is therefore exposed to erosion. The pressure of the heavy machines in use in-
creases the risk of soil tightening which has also been found in Finnish sugar beet fields (ERJALA & RAININKO, 1985; RAININKO, 1988). Soil microorganisms which form soil aggregates have favourable effects on the soil structure and microorganisms of sugar beet soil are therefore particularly important. The microorganisms are also the most important factors for the decomposition and forming of nutrients available for plants and in the degradation of pesticides which could leave residues.

The effects of individual pesticides as well as some pesticide programmes on the soil biology of sugar beet fields have been studied (PESTEMER & MALKOMES, 1983), but the pesticides used in these experiments are not used in Finland. The aim of our study was to determine the possible microbial response to pesticides in Finland. The pesticide doses for the present study were selected to correspond a) to the normal, recommended dose and b) to 150% of this dose, because slight overdoses (or underdoses) of pesticides may be typical resulting from uneven distribution or broken spraying machines, which is rather common in Finland (LUOMA & LAVONEN, 1987).

Seed dressing compounds thiram and hymexazol are fungicides. This mixture has been found to control effectively the damping-off of sugar beet (VESTBERG et al. 1982).

Hymexazol is known to affect, in particular, *Fusarium*, *Aphanomyces*, *Pythium*, *Corticium* (PESTICIDE MANUAL, 1979) Mortierella, some *Phytophthora* (TSAO & GUY, 1977), *Alternaria*, Botrytis, and *Phoma betae* (VURBANOV et al. 1984). The effects of hymexazol on non-target soil microbes have been described very sporadically. Hymexazol only in high concentrations inhibits *Saccharomyces*, *Pseudomonas* (KAMIMURA et al. 1976), red clover rhizobia (HEINONEN-TANSKI et al. 1982) and various phytopathogenic bacteria (TOMITA et al. 1975; OROS et al. 1983). The Bengal gram rhizobia studied by GARG et al. (1981) were more sensitive to hymexazol than the above mentioned microbes. Hymexazol is degraded by soil microorganisms to carbon dioxide, acetoacetamide and oxazolone (NAKAISHI et al. 1974).

As a fungicide, thiram is known to reduce the fungal activity of soil (ANDERSON et al. 1981; HICKSCH et al. 1984) and the numbers of some bacteria, e.g. nitrogen-fixers (reviewed by TORSTENSSON, 1979). Thiram application increases the number of phosphate mobilizing microbes in soil (WAINWRIGHT & SOWDEN, 1977).

The effect of dimethoate on soil microbes is poorly known, but may be short-lived (CONEGADO et al. 1979) owing to the relatively rapid degradation of dimethoate (BROBSMUSEN et al. 1969).

Phenmedipham in normal doses decreases the biological activity of some sugar beet soils for a short time (BELLINCK & MAYAUDON, 1978; VERSTRAETE et al. 1979) and inhibits the number of *Azotobacterium* (SIMON-SYLVESTRE & BEAUMONT, 1982). On the other hand, it increases cellulose degradation and ammonification (SIMON-SYLVESTRE, 1979). The effects of phenmedipham on nitrification may be inhibitory or stimulatory, depending on soil type (SIMON-SYLVESTRE, 1979; VERSTRAETE et al. 1979). The oxidation of ammonia to nitrite is less sensitive than the oxidation of nitrite to nitrate (RATNAYAKE & AUDUS, 1978), which can result in the enrichment of nitrite in soil. Excessive concentrations of phenmedipham strongly inhibit soil nitrification (TENA et al. 1984).

As a general conclusion, the effects of phenmedipham are less negative on soil microorganisms than those of metamitron or some other herbicides used with sugar beet (VERSTRAETE et al. 1979). Metamitron can stimulate urease activities in soil, but it has only minor effects on phosphatase, on the numbers of various microbial groups (VOETS et al. 1977; GADKARI, 1984), and on nitrogen and carbon transformation (MALKOMES, 1987). The nitrogen fixation of some cyanobacteria is inhibited by metamitron (GADKARI, 1987).

Alloxidim-Na degraded microbologically rapidly in two sandy soils at 25°C (ONO et al.
1984). The half-lives were approximately 5—6 days. Many unidentified and identified metabolites (including CO₂) were found. In Swedish experiments, however, alloxidim-Na could still be found up to 2—3 months after the spring spraying (Nilsson, 1984). The persistence was higher in northern Sweden than in southern Sweden. Alloxidim-Na has a slight inhibitory effect on pea nodulation (Bebb et al. 1985), but no effect on the degradation of herbicide benazolin (Kostowska et al. 1982).

Materials and methods

The sugar beet (Salohill) has been cultivated in loam containing 10.7 % organic matter in Laukaa (62° 28’ N, 25° 56’ E) and in silty clay containing 8.7 % organic matter in Perniö (60° 17’ N 23° 7’ E) in 1982—1985. The seeding (10⁵ seeds/ha) was done in Perniö by sown-to-stand by 15 cm seed spacing with simultaneous fertilizing (EriJala & Raininko, 1985; Raininko, 1988).

The soils are described in Table 1. The trials were carried out in four parallel plots (9 m × 12 m in Laukaa and 14.4 m × 36 m in Perniö). The control plots were not dosed with pesticides, the weeds were hand-weeded in the second weeks of June and July. The pesticides were applied at normal recommended dosages or 150 % of the normal dosages. The agricultural operations and sampling schedules were

Table 1. Properties of the soils at the beginning of the trials.

<table>
<thead>
<tr>
<th></th>
<th>pH</th>
<th>K mg/l</th>
<th>P mg/l</th>
<th>Mg mg/l</th>
<th>Ca mg/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laukaa</td>
<td>6.2</td>
<td>108</td>
<td>14</td>
<td>142</td>
<td>1708</td>
</tr>
<tr>
<td>Perniö</td>
<td>6.7</td>
<td>241</td>
<td>21</td>
<td>614</td>
<td>422</td>
</tr>
</tbody>
</table>

<p>| | | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Laukaa</td>
<td>Sand %</td>
<td>Silt %</td>
</tr>
<tr>
<td>Laukaa</td>
<td>30</td>
<td>43</td>
</tr>
<tr>
<td>Perniö</td>
<td>17</td>
<td>34</td>
</tr>
</tbody>
</table>

Table 2. Agricultural schedule and sampling times.

<table>
<thead>
<tr>
<th></th>
<th>Control plots</th>
<th>Normal pesticide dose</th>
<th>150 % of the normal pesticide dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>May</td>
<td>sampling</td>
<td>sampling</td>
<td>seeding + seed dressing with 6.8 g/ha thiram + 17.5 g/ha hymexazol²</td>
</tr>
<tr>
<td>Late May</td>
<td>seeding</td>
<td>450 g/ha phenmedipham + 240 g/ha dimethoate</td>
<td>675 g/ha phenmedipham + 360 g/ha dimethoate</td>
</tr>
<tr>
<td>Early June</td>
<td>—</td>
<td>450 g/ha phenmedipham + 2800 g/ha metamitron + 240 g/ha dimethoate</td>
<td>675 g/ha phenmedipham + 4200 g/ha metamitron + 360 g/ha dimethoate</td>
</tr>
<tr>
<td>Middle of June</td>
<td>sampling</td>
<td>471 g/ha phenmedipham + 1500 g/ha alloxidim-Na³</td>
<td>707 g/ha phenmedipham + 2250 g/ha alloxidim-Na³</td>
</tr>
<tr>
<td>Late June</td>
<td>—</td>
<td>sampling</td>
<td>sampling</td>
</tr>
<tr>
<td>Early July</td>
<td>—</td>
<td>sampling</td>
<td>sampling</td>
</tr>
<tr>
<td>Early Aug.</td>
<td>sampling</td>
<td>sampling</td>
<td>sampling</td>
</tr>
<tr>
<td>Early Sep.</td>
<td>sampling</td>
<td>harvesting</td>
<td>harvesting</td>
</tr>
<tr>
<td>Oct.</td>
<td>harvesting</td>
<td>harvesting</td>
<td>harvesting</td>
</tr>
</tbody>
</table>

¹ 150 % of the normal dosage in Laukaa  
² not in 1982 in Laukaa  
³ alloxicim-Na only in Laukaa
done as needed; they are presented in Table 2. In some cases dimethoate and a second herbicide treatment with phenmedipham and metamitron were applied before the second sampling. Alloxidim-Na was used only in Laukaa. No hymexazol was used in 1982 in Laukaa.

The sub-samples, to a depth of 3 cm, were collected from five parts of the plots, bulked and mixed. The samples were never taken from the area within 1 m of the border of each plot. The microbiological inoculations were made on the sampling day. The soils were kept at 5°C for the next day for the dehydrogenase determinations. For the determination of the nitrification activity, the soil samples were air-dried at room temperature for 3—10 days.

The number of “total” microorganisms were determined on Taylor’s (1951) agar. The numbers of spore-forming aerobes were determined on agar of Fenchel and Hemmingsen (1974) after destroying the non-sporing microorganisms by heat-treatment at 80°C for 20 min. The ureolytics were determined by the MPN-technique in the Christensen (1946) medium. The incubations were kept at 15°C for 3—4 weeks.

The determination of dehydrogenase has been described by Mettala et al. (1982) and the nitrification by Heinonen-Tanski et al. (1985). The cellulose decomposition was determined by the polyester-bag method, following the weight loss of filter paper (5 g, Schleicher & Schüll 604) buried to a depth of 2 cm for three to four months. The papers were washed carefully, dried at room temperature and weighed.

The t-tests and paired t-tests were performed by using natural log transformations of the microbial groups at all the sampling times (14) and both of the places (2) and treatments (2).

Results

The soil microbial results are presented in Table 3, giving the means for the microbial numbers and activities during the entire experimental time. The most significant statistical differences in Perniö were found for de-

<table>
<thead>
<tr>
<th></th>
<th>Laukaa experiment</th>
<th></th>
<th></th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Control plots</td>
<td>Normal pesticide</td>
<td>150 % of the normal</td>
</tr>
<tr>
<td>Total number $\times 10^7$</td>
<td>41</td>
<td>36</td>
<td>43</td>
</tr>
<tr>
<td>Spore-forming aerobes $\times 10^4$</td>
<td>83</td>
<td>85</td>
<td>70</td>
</tr>
<tr>
<td>Ureolytics $\times 10^4$</td>
<td>77</td>
<td>57*</td>
<td>52**</td>
</tr>
<tr>
<td>Dehydrogenase TPF $\mu$g/g</td>
<td>80.6</td>
<td>73.8*</td>
<td>74.1</td>
</tr>
<tr>
<td>Nitrification NO$_3$-N $\mu$g/g</td>
<td>73.8</td>
<td>77.6</td>
<td>71.5</td>
</tr>
<tr>
<td>Cellulolysis %</td>
<td>40.5</td>
<td>41.9</td>
<td>40.2</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Perniö experiment</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control plots</td>
<td>Normal pesticide</td>
<td>150 % of the normal</td>
</tr>
<tr>
<td>Total number $\times 10^7$</td>
<td>22</td>
<td>20</td>
<td>25</td>
</tr>
<tr>
<td>Spore-forming aerobes $\times 10^4$</td>
<td>80</td>
<td>81</td>
<td>76</td>
</tr>
<tr>
<td>Ureolytics $\times 10^4$</td>
<td>12</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Dehydrogenase TPF $\mu$g/g</td>
<td>51.5</td>
<td>46.8*</td>
<td>46.0**</td>
</tr>
<tr>
<td>Nitrification NO$_3$-N $\mu$g/g</td>
<td>119</td>
<td>126</td>
<td>112</td>
</tr>
<tr>
<td>Cellulolysis %</td>
<td>36.2</td>
<td>34.3</td>
<td>32.0</td>
</tr>
</tbody>
</table>

* P<0.01 and ** P<0.05
hydrogenase activity (P = 0.021 in paired t-tests for the plots with normal pesticides and P = 0.009 for the plots with 150 % pesticide dosage and in Laukaa for dehydrogenase activity (P = 0.036 for plots with normal pesticide dosage) and for ureolytics (P = 0.035 for the normal pesticide dosage and P = 0.006 for the 150 % pesticide dosage).

The differences between microbial numbers or activities during separate experiments and sampling times were statistically significant (P < 0.05) in some cases (56 cases together in both places and treatments). In these cases the microbial numbers or activities of the pesticide-treated plots were lower than in the controls nine times in Laukaa and ten times in Perniö, and higher three times in Laukaa and twice in Perniö. Both the normal doses and the overdoses caused these effects. The effects were temporary. The dehydrogenase activity tests differed significantly eleven times and the number of the ureolytics five times.

The sugar beet yields in Laukaa and Perniö are presented in Table 4. The beet and sugar yields in plots with normal the pesticide dosage were higher than the control although the difference was not statistically significant. The yields in the plots with the 150 % pesticide dosage were similar or less than in the plots with normal pesticide dosage in Perniö but similar or higher in Laukaa. The dry weights of top yields were measured only in Laukaa. The top yields were statistically significantly (P < 0.05) higher in pesticide-treated plots than in the controls. No symptoms of damage caused by pesticides or diseases were found.

**Discussion**

The degradation of metamitron used in relatively high doses depends very much on temperature. A decrease of 10°C retarded the degradation to 1/4 of the degradation rate at higher temperature (Fuhr & Mittelstaedt, 1979), whereas a few warm days did

<table>
<thead>
<tr>
<th>Year</th>
<th>Beets (t/ha)</th>
<th>Sugar (kg/ha)</th>
<th>Top dm (kg/ha)</th>
<th>Beets (t/ha)</th>
<th>Sugar (kg/ha)</th>
<th>Top dm (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laukaa experiment</td>
<td>Normal pesticide</td>
<td></td>
<td>150 % of the normal</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1982</td>
<td>2.0</td>
<td>308</td>
<td>500</td>
<td>5.8</td>
<td>481</td>
<td>1 350</td>
</tr>
<tr>
<td>1983</td>
<td>33.1</td>
<td>6 424</td>
<td>3 090</td>
<td>40.2</td>
<td>7 833</td>
<td>5 580</td>
</tr>
<tr>
<td>1984</td>
<td>9.2</td>
<td>n.d.</td>
<td>1 450</td>
<td>8.0</td>
<td>n.d.</td>
<td>2 090</td>
</tr>
<tr>
<td>1985</td>
<td>24.5</td>
<td>n.d.</td>
<td>4 260</td>
<td>30.5</td>
<td>n.d.</td>
<td>6 790</td>
</tr>
<tr>
<td>1986</td>
<td>22.5</td>
<td>n.d.</td>
<td>3 170</td>
<td>20.2</td>
<td>n.d.</td>
<td>4 670</td>
</tr>
<tr>
<td>100 (100)</td>
<td>115 (129)</td>
<td>165</td>
<td>129 (129)</td>
<td>187</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Year</th>
<th>Beets (t/ha)</th>
<th>Sugar (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Perniö experiment</td>
<td>Normal pesticide</td>
<td></td>
</tr>
<tr>
<td>1982</td>
<td>33.2</td>
<td>5 169</td>
</tr>
<tr>
<td>1983</td>
<td>44.3</td>
<td>7 262</td>
</tr>
<tr>
<td>1984</td>
<td>16.3</td>
<td>2 578</td>
</tr>
<tr>
<td>1985</td>
<td>25.3</td>
<td>4 160</td>
</tr>
<tr>
<td>100</td>
<td>107</td>
<td>106</td>
</tr>
</tbody>
</table>

Table 4. Sugar beet (t/ha) and sugar yields (kg/ha) and Laukaa dry matter of tops (kg/ha) in control plots, plots with the normal pesticide dosage and plots with 150 % of normal pesticide dosage. dm = dry matter. n.d. = not determined.
not markedly increase the degradation of metamitron. Thus Führ and Mittelstaedt (1979) assumed that there is practically no degradation of metamitron in the German climate from November to March. In the climatic conditions of Finland, the degradation of metamitron may be even slower and limited to a shorter period. Allen and Walker (1987) also found that the degradation of metamitron depends positively on the sand percentage and negatively on the silt and clay percentages of soil. Thus we can assume that the degradation of metamitron may have taken place more rapidly in Laukaa than in Perniö due to the coarser soil texture in Laukaa. The residues of metamitron (not measured) may still have been considerable, possibly reducing the sugar and sugar beet yields, especially in plots administered 150 % of the normal herbicide doses in the Perniö experiments. Also the sugar percentage was reduced in Laukaa in pesticide-treated plots in 1983; the sugar yields of 1982 were so low in Laukaa that conclusions are difficult to draw. Unfortunately, the sugar content was not measured lately in the Laukaa experiments.

Alloxidim-Na may have degraded well also in the Laukaa experiments giving CO₂ and the free and humus-bound residual degradation products found in Japanese outdoor experiments (Ono et al. 1984), although this may not have occurred in a few days but in a few weeks or months, as in Sweden (Nilsson, 1984).

The normal application of pesticides had a favourable effect on sugar beet and sugar yields in this trial. But the overdoses of pesticides, 150 % of the normal gave only the same or slightly lower sugar yields than the normal doses during the years tested. According to this result overdoses of sugar beet pesticides, especially herbicides, should be avoided.

All the statistically significant effects caused by pesticides on soil microorganisms, both stimulative or inhibitive, must be considered to be unfavourable because they disturb the dynamic equilibrium of the soil microorganisms.

Often these effects can be regarded as a symptom of overly long-lasting residues of pesticides or their degradation results. Many pesticides with "unusual" chemical structure, e.g. with halogenation, aromatic or triazine rings, are degraded by cometabolism. Cometabolic degradation requires another carbon source and is the joint work of many microorganisms. Thus the degradation of metamitron is cometabolic according to the results of Führ and Mittelstaedt (1979) and Engelhardt et al. (1982). The cometabolic degradation can be accelerated by increasing the general microbial activity (Torstensson & Stenström, 1986). The general microbial activity can be increased, for instance by adding reasonable amounts of mineral fertilizers or by application of crop rotation (Mettälä et al. 1982), or by adding organic compounds (Campbell, 1985). Rotation with ley caused a very great increase in microbial activity in Finnish clay soil (Heinonen-Tanski, 1986).

To be sure of the high general microbial activity necessary also for the degradation of pesticides and for good soil structure, knowledge is needed about practical methods for increasing microbial activity. The possible methods could be either the use of sugar beet tops and other organic matter as fertilizers or suitable and economical rotation.

Acknowledgements. We wish to thank Ms. Pirjo Halonen M. Sc. for statistical calculation of the microbial results, Ms. Sevastiana Ruusamo M. A. for correcting the English language and many students for their microbial laboratory work.
References


CHRISTENSEN, W.B. 1946. Urea decomposition as a means of differentiation of Proteus and paracolon cultures from each other and from Salmonella and Shigella types. J. Bact. 52: 461—466.


ous placement of fertilizer at drilling in one operation to save costs and increase yield. I.I.R.B. 51st Winter Congress Proc. 23—32.


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SELOSTUS

Sokerijuurikkaan viljelyssä käytetyn torjuntaaineohjelman vaikutus maan mikrobeihin ja sokerijuurikkaan satoihin

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Sokerijuurikassadot lisääntyivät torjunta-ainekäsittelyllä, mutta yliannostus ei enää lisännyt satoa normaali-käsittelyyn verrattuna. Perniön kokeessa sokerisato oli yliannoksen saaneissa ruuduissa pienempi kuin normaaliannoksen saaneissa. Täten satotoiveita ajatellen ei ole

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mitään syyä ylisuurien torjunta-aineiden käyttöön. Ta-hattoman yliannostuksen välttämiseksi ruiskujen kuntoon olisi kiinnitettävä huomiota, erityisesti herbisidien koh-dalla.


Olisi syytä selvittää jatkossa keinoja, joilla sokerijuu-rikasmaiden yleistä mikrobiologista aktiivisuutta voitai-siin kohottaa. Mahdollisia keinoja voisivat olla organi-set aineet (naatit, kohtuullinen määrä olkia ym.) sekä eri-laiset viljelykierrot sisältäen mahdollisesti myös nurmen.