Investigation of the relationship between the level of ALKALOIDS and degree of PEST DAMAGE in *LUPINUS POLYPHYLLUS* LINDL.

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Work by Boguslav Kurlovich presented by David McNaughton
Introduction and Object

The first sweet forms of lupins were created almost 90 years ago by Reinhold von Sengbusch in Germany. The Russian Institute of Plant Industry (VIR) also developed its own research as well as an instruction to identify alkaloid-free lupins. It was immediately published with the foreword by N.I. Vavilov (Ivanov et al., 1932). Several thousand plants were analyzed by this method and the first sweet form of L. polyphyllus was bred in 1932 at VIR. The above-mentioned publication of VIR as well as the discovery of German scientists was a cornerstone of the modern breeding work with low-alkaloid fodder (sweet) lupins in the whole world. We produced the first fodder commercial variety of L. polyphyllus Lindl. named ‘Pervenets’ (the first sweet variety) for the conditions of the Northwest Russia (Kurlovich, 2002). Variety ‘Truvor‘ was developed for the conditions of Ukraine. Now we are conducting research on sweet forms of the perennial Washington lupin in Finland, New Zealand and UK. We present some of our research results in this paper. We noticed that low alkaloid plants are exposed to pest. This makes it easy to detect sweet plants, but they are difficult to replicate and keep from pests. The developed approaches may also be applied to other cross-pollinated lupin species, such as L. mutabilis Sweet., L. nootkatensis Donn., L. arboreus Sims., L. perennis L., L. elegans H.B.K., L. hartwegii Lindl. and other forms promising for agricultural production.

Object: Accessions of Lupinus polyphyllus with different alkaloid content from the collection of the N.I.Vavilov Institute of Plant Genetic Resources (VIR), forms of lupin with different alkaloids from Finland and samples with different levels of alkaloids from New Zealand and UK.
Materials and Methods

- Qualitative evaluation of alkaloid activity was carried out by a conventional traditional method with the help of test paper impregnated with Dragendorf’s solution.
- Gas chromatography was used for the quantitative analysis of alkaloids. The extraction and analyses were conducted following the procedure presented by Kamel et al. (2016).
- Part of the analysis is performed by Prof. Vito Boido from the laboratory of Pharmacology and Neuroscience, National Institute for Cancer Research, Genoa, Italy.
- We used the scale of the descriptor list for lupin while measuring the degree of plant susceptibility to pests: 1 – No symptoms or very weak (< 2.5%), 3 – Slight (2.5-10%), 5 – Intermediate (11-25%), 7 – Severe (26-50%), 9 - Very severe (>50%).
The most harmful Pests of *Lupinus polyphyllus* Lindl. in conditions of Finland
The relationship between plant susceptibility to pests and the level of alkaloids (average for 3 years)

<table>
<thead>
<tr>
<th>SUSCEPTIBILITY TO PESTS</th>
<th>ALKALOIDS, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>No symptoms</td>
<td>4.00</td>
</tr>
<tr>
<td>Slight</td>
<td>2.00</td>
</tr>
<tr>
<td>Intermediate</td>
<td>0.20</td>
</tr>
<tr>
<td>Severe</td>
<td>0.09</td>
</tr>
<tr>
<td>Very Severe</td>
<td>0.02</td>
</tr>
</tbody>
</table>
The relationship between plant susceptibility and level of alkaloids
The leaf evacuated by pests with a residual alkaloid content of 0.02-0.04% (from left) in comparison with not damaged leaf.
We noticed that low alkaloid plants are exposed to pest. This makes it easy to detect sweet plants. The degree of damage of the plant by pests can be the marker that allows to identify low alkaloidal (sweet) forms of lupins. Strongly affected plants are usually low alkaloidal. This marker will significantly accelerate and simplify the search for low-alkaloid forms. In this case, there is no need to test for alkaloids a huge number of plants.

On the other hand these plants are difficult to multiply and keep healthy and not damaged. Which is more important of these two conclusions? Tolerance to diseases and pests is very important for lupins. However, the lack of alkaloids in the plant reduces its tolerance.

We believe that our observations will contribute to the development of new low-alkaloid (sweet) varieties of different American lupin species (subgen. *Platycarpos* (S. Wats.) Kurl.).
Conclusions

The Express method of finding of low alkaloid plants of lupins is offered. The degree of damage of the plant by pests can be the marker that allows to identify low alkaloidal (sweet) forms of lupins. Strongly affected plants are usually low alkaloidal.

The negative link between low alkaloid and pest resistance has been revealed. Low alkaloidal plants are exposed to pest. This phenomenon should be considered in the breeding.
We thank colleagues for the help!

- We are grateful to the professor Vito Boido (Laboratory of Pharmacology and Neuroscience, National Institute for Cancer Research, Largo R. Benzi, 10, 16132, Genoa, Italy), who has performed a significant part of the analyses on quantitative definition of alkaloids in *Lupinus polyphyllus* Lindl.
Soya UK Work with Polyphyllus
2015 - 2019

- 2015 – Met Gavin Loxton at conference in Milan, and subsequently agreed to begin testing perennial low-alkaloid lupins in the UK.

- 2016 – Seed from New Zealand was spring planted with 1.5kg of seed being sown on an area of 36 metres x 36 metres. Seed was inoculated with Legume Technology commercial bradyrhizobium lupin inoculant.

- Emergence painfully slow and inconsistent. Acid treatment a possibility in future?

- Spot-sprayed with glyphosate for weeds, and also blanket sprayed with 24 metre commercial boom sprayer for grass weeds.
Soya UK Work with Polyphyllus 2015 - 2019

- 2017 – Kept killing the weeds and observing.
- Harvest 2017 – collected 5kg of seed from 36x36m plot.
- Spring 2018 – planted 0.8 Ha with a mixture of Polyphyllus and Iris blue lupin.
- All seed inoculated and whole plot was sprayed with pre-emergence mix of Imazamox + Pendimethalin.
- Blue lupins harvested in August with newly established Polyphyllus still in vegetative stage. Seed plant removed any stray seeds, so pure crop of Iris was harvested.
- Remaining crop of Polyphyllus was left to mature and then heavy rolled to incorporate seeds into the soil surface in Autumn 2018
Soya UK Work with Polyphyllus 2015 - 2019

- Spring 2019 – plan to roll the 0.8 Ha again, and then treat with Lentagran (Pyridate) and a commercial graminicide to kill the weeds. Need to assess any negative effect from the chemicals.
- Summer 2019 – plan to graze the crop with fattening lambs, and use dragendorff paper to test and remove any plants that are not enthusiastically eaten. Need to see safety of feeding to sheep – will try not to kill any and will provide back-run to grass.
- Do proper assessment of nodulation.
- Also plan to use gas chromatography to do alkaloid analysis in 2019 /2020.
Soya UK Work with Polyphyllus 2019 Onwards?

Spring 2020 Onwards?

Who knows?

Stick with 0.8Ha
See what happens
See if it is possible to obtain measurable grazing results
See if the alkaloids are stable and safe
Test more herbicides
Assess nodulation & guess at total N-fixation for companion crops.

At some point - take a commercial view on UK viability
Thank You / Gracias

David McNaughton
Soya UK Ltd