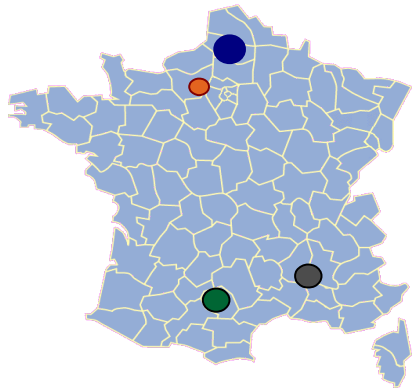




***Lunaria : culture and oil production for
bio-lubricants and nutraceutical industry***



Company's PRESENTATION

S.M.E. based in Picardy (80)
French Production on 11 000 Ha
120 employees on 6 locations, 20 M € CA

Airaines - Head Office - Machine : conditioning.
Breeding, Seed Multiplication and Production.
Quality & Analysis Laboratories, Certification...



Tourny - Subsidiary **TIWY** - Specialized
in nutrition for birds and rodents.

Lavour & Montélimar - Breeding, production.

Argentina : winter-season - sunflower, sorghum, linseed...



Verona : ITALY - Subsidiary **SEMFOR** ; Commercialization
of fodder crops, maize, linseed, pea, sorghum...

RESEARCH and DEVELOPMENT

SEEDS PRODUCTION

Fodder Crops, Grass Graminaceae :

English, italian and hybrid **rye-grass**

Dactyl ; **Red and tall fescue**

Leguminous plants :

White, purple and sub-clover

Common vetch ; Alfalfa



Laboulet has 20% of french fodder crops and grass market.

RESEARCH and DEVELOPMENT

BREEDING PROGRAM

Sunflower : oleic (biodiesel)

Linseed : industry, Omega 3

Rapeseed : HEAR, HOLL, 00

Lunaria

Maize

Sorghum

Protein pea



LABOULET's Assets :

UNIQUE and POWERFUL VEGETAL MATERIAL

ASSOCIATION of COMPETENCES

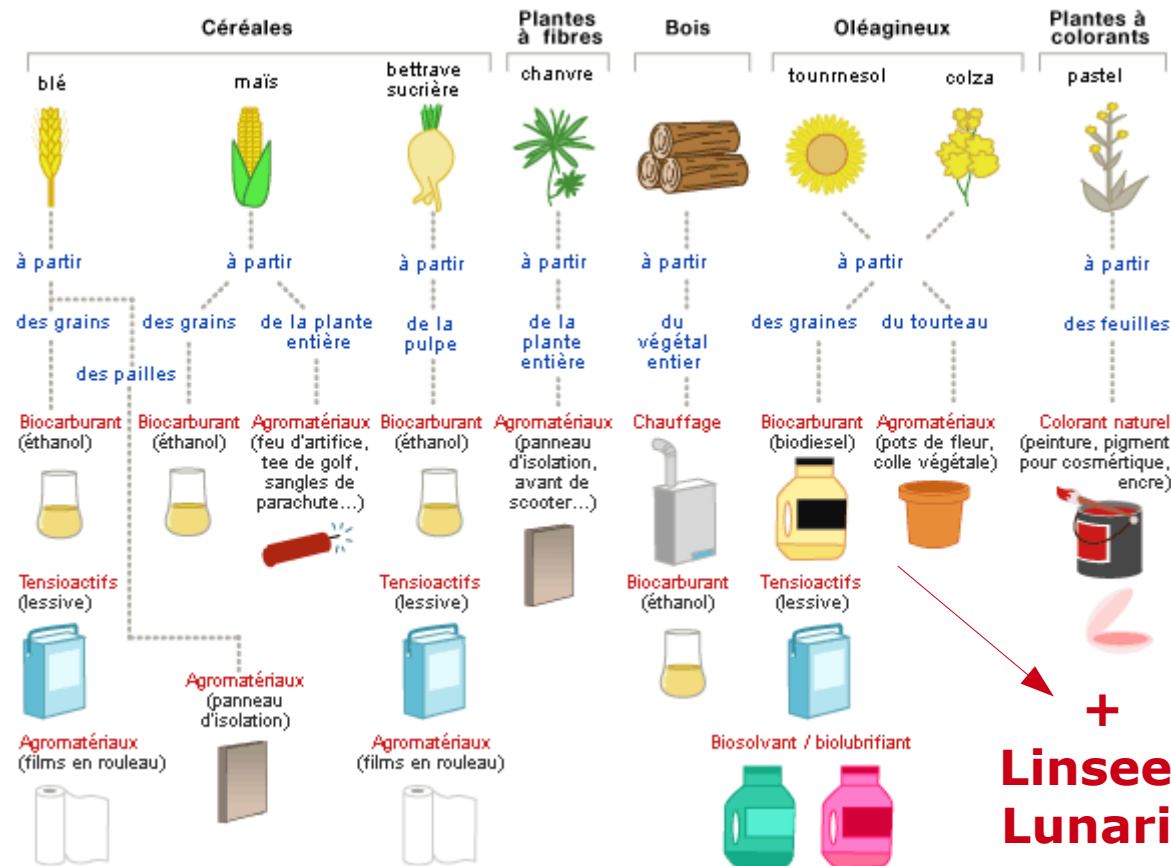
Active Partner of Competitiveness Pole « AGRO-RESSOURCES and INDUSTRIES »

PERFECT ADAPTATION to FARMERS and END-USERS needs



Supply of raw bio-products

Plants represent a **significant source of agricultural resources, renewable raw materials** and alternatives to replace petroleum, enabling the manufacture of **bio-based products** - surfactants, solvents... The opportunities are numerous, significant and high value added, including:



Source : environnement-online.com



+
Linseed Lunaria



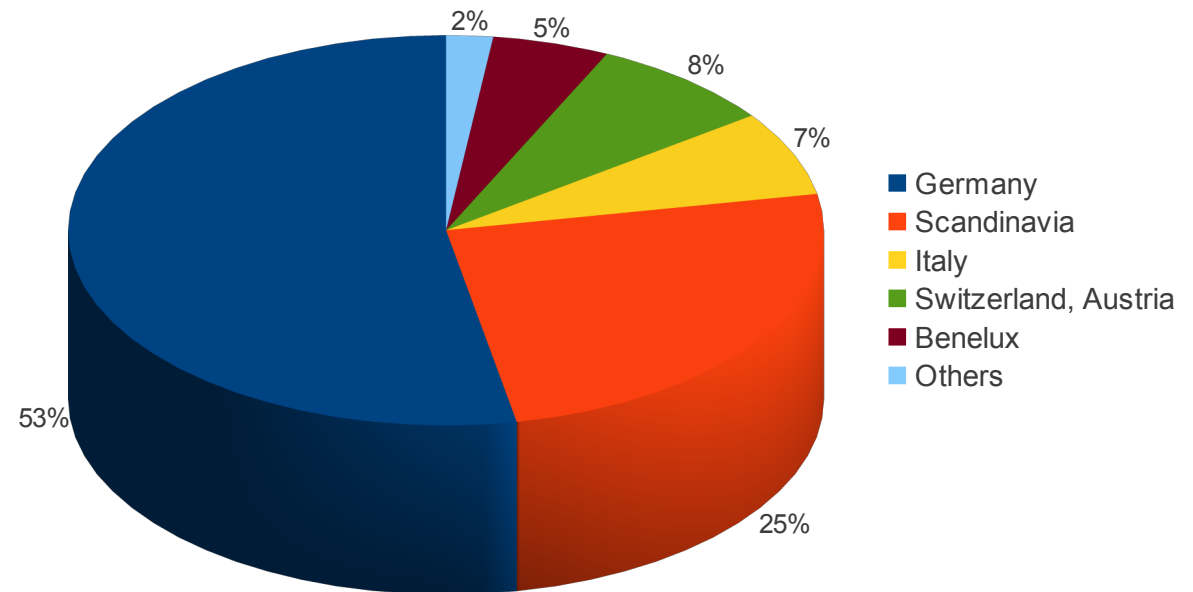
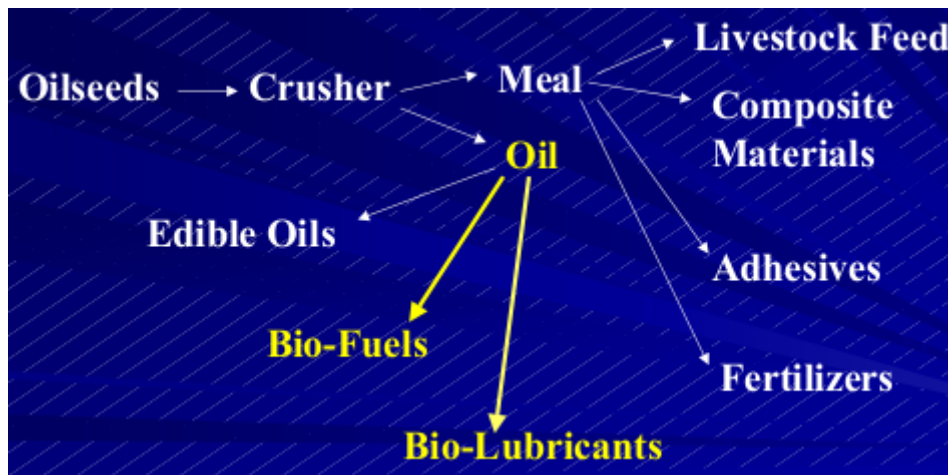
Biolubricants market in Europe

Lubricants market
 Europe 5 000 000 t
 France 850 000 t

Biolubricants production
 Europe 100 000 t
 Germany 50 000 t
 France 1 000 t

Today : bio-lubricants = 2 % of market, but increasing.

Potential : 200 000 t in 2015



Use of lubricants

In metals and greasing work, lubricant have several functions :

- decrease the efforts and energies of working
- reduce the friction coefficient
- reduce the metal's heating
- obtain the required surface quality (shining, smooth...)
- avoid corrosion
- preserve the product...



European Commission decisions :

- **Environmental & human preservation, grounds and water quality, Reach, Ecophyto 2018...**
- **Elimination of waste mineral oils**
- **Prohibition of use of additives containing chlorinated and sulphur substances**

These additives were used to increase the lubrication potential, there are currently replaced by esters of plants, and **Lunaria could be a perfect candidate, due to its lubricating properties.**

The concept of regeneration, recyclability of the products, pollution of the grounds gives an opportunity to the formulations based on biodegradable and non-toxic vegetable oil.



Lubricants - Vegetable Oils

To answer the constraints, industrialists of metal's work use cutting oils in two forms:

- Straight cutting oils : mix of mineral oil 95% and additives
- Soluble oils : mix of 93-97 % of water and 3-7 % of formulated mineral oil, composed from 40-75 % of oil, 15-20% of emulgator, remainders are additives (of lubrication, anti-corrosive, antimicrobial...)

If performances of these soluble mineral oils are excellent, they cause in the same way many legislative and technical problems:

- the formulations are elaborated from mineral oil (CO_2)
- additives causes environmental toxicity
- The obstruction of these recovering units of water and worn oils is limiting
- costs of worn water recycling
- conservatives make the fluids allergens

The current tendency is the development of new generations of cutting oil:

- **Micro lubricants**
- **Greases**
- **Cutting oil based on vegetable oil**

Mineral oil replaced by vegetable oil & rapeseed / sunflower esters, but in these formulations: problems

- Capacity of vegetable oils lubrication
- Availability of emulgator adapted to vegetable oils
- Oxidation stability

LUNARIA

Lunaria Annua L.

Botanical classification

Common names: Honesty (English), Monnaie du pape (french)

Synonym: *Lunaria biennis* - Class: Magnoliopsida - Dicotyledons

Family: Brassicaceae - Mustard family (alt. Cruciferae)

Genus: Lunaria L. - Species: Lunaria annua L. - Annual honesty

Origin : native of south east Europe and western Asia.

Description

Lunaria annua L. is a biennial cruciferous garden flower which produces characteristic silver, translucent seed silicula ('Silver Penny'). Found commonly along roads, in semi-shady places. Has an ornamental value.

Height : 40 to 100 cm - Stem : stiffly hairy - Leaves : cordate-acuminate, coarsely toothed, roughly hairy, lower ones long-stalked

Flowers purple / white, 4 petals - Seedpods large, circular, strongly set to stem, 6 to 10 flat seeds compressed in 2 rows, TKW 15-20 gr



LUNARIA

- High content in **erucic** C 22, and **nervonic** acid
- Replace mineral oils by **vegtal oils** in Europe
- Markets : **bio-lubricants, nutraceutic**

	<i>Linseed</i>	<i>Erucic rapeseed</i>	<i>Lunaria</i>
Vegetable production	25 qx / Ha	35 qx / Ha	15-20 qx / Ha
Oil Content	46%	48%	30 - 40%
C22 content	0%	45%	40-45%
C24 content	0%	1%	20-25%



LUNARIA

Erucic acid : biodegradable, high tolerance to T°, easy polymerization and drying, high calorific value & cetane number

Use : lubricants (transmission oil), binder for oil paints, cosmetics, manufacture of emulsions (photographic films), surfactants, emollients (healthcare products), plastics, nylon, polyester, bio-diesel...

Nervonic acid : mono-unsaturated Omega-9 f.a. Found in sphingolipids of white matter human brain, role in biosynthesis of **nerve cell myelin**, constituent of membrane phospholipids.

Used in treatment of disorders involving demyelination – adrenoleukodystrophy, multiple sclerosis, premature birth.

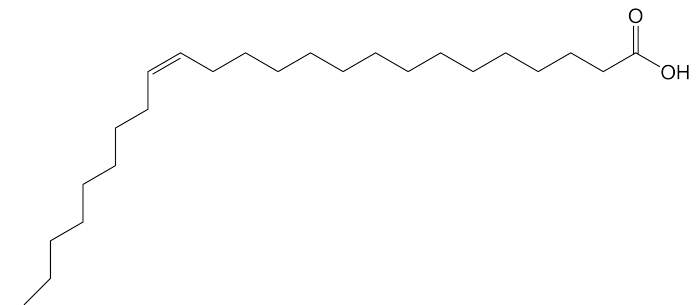
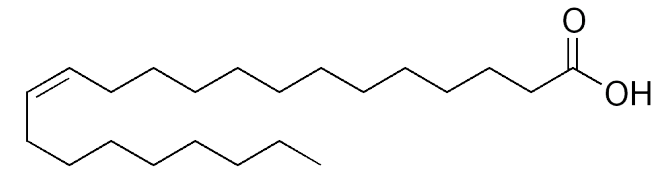
Nervonic acid and derivatives : ability to be produced from lunaria oil by conventional techniques. Use in pharmaceuticals.

Products containing lunaria oil are in clinical development.

Effects of feeding Lunaria oil rich in nervonic and erucic acids on the fatty acid compositions of sphingomyelins from erythrocytes, liver, and brain of the quaking mouse mutant

Claire Cook, Joan Barnett, Keith Coupland and John Sargent, Lipids - 1998, Vol. 33, N° 10, pp. 993-1000

Feeding an oil from Lunaria biennis rich in 22:1n-9 and 24:1n-9 to homozygous quaking mice caused a large increase in the percentage of 24:1n-9 and corresponding decreases in the percentage of 24:0 and 22:0 in sphingomyelins from liver, erythrocytes, and milk.



LUNARIA

Lunaria Annua L.

Oil

- High lubricant capability
- High smocking point
- High mixing properties
- Good viscosity

Laboulet's lunaria variety :

Coefficient of Friction *Average SCAR X/Y*

Colza 00	9.2	180
Erucic rapeseed	8.8	154
Lunaria	6.8	132
Jojoba	9.7	177
Mineral oil	8.7	250

Coefficient of friction and wear are low.

This is very positive for lubricant application.



LUNARIA

Agronomy – Main characteristics

Positive characteristics of *Lunaria annua*

- Low chemical input requirement
- Low dehiscence even after maturation
- Simple mechanization
- No special processing required for oil & meal quality
- Honesty is well adapted to a temperate climate

Negative characteristics of *Lunaria annua*

- Limited knowledge of crop culture & potential
- Limited germplasm still to be improved
- Rusticity and adaptability
- Pest and diseases : Alternaria, Albugo candida
Beetles (*Meligetis aeneus*)
Powdery mildew



Biennial character of Lunaria = main constraint for an economically profitable oil production.

LUNARIA

Agronomy – Lunaria annua

Biennial lunaria : long juvenile phase, must be sown early in summer to achieve vigorous plant development, necessary because of high vernalisation requirement.

But : low yields, prohibitive end price.

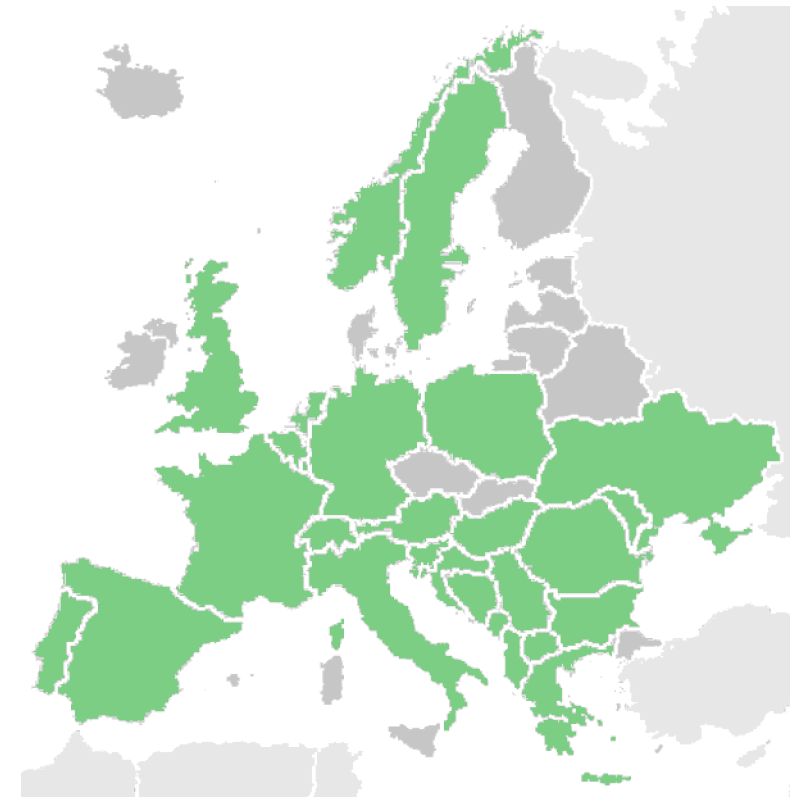
Alternative : open seeded crop over 18 month period, with easier inter-row weed control. End product price remains too expensive.

Sowing into a standing crop of winter wheat, with a special seed-drill, in mid-March : establish & thrive underneath growing cereal. Wheat is harvested in August, leaving young lunaria plants to grow away from within the stubble.

This solution allows two distinct monetary returns over the 2 years period : first from winter wheat, second from undersown lunaria.

=> Laboulet's work : development of a **late winter annual type** which can be sown later in summer after an early harvested crop.

Material from Netherlands, Switzerland... Cultivation, breeding.



LUNARIA

Challenge : industrial production of *Lunaria*

Work with an annual variety (*sow. end August North Fce*)

Production potential and agronomy of *Lunaria* crop required lots of investigation :

- Control of crop management, improvement and stabilization of yields (grain, oil)

Ground's choice, sowing date and density, herbicides, fungicide, insecticide, fertilization N-P-K-S, harvest...

- **Optimization of production conditions**

Experiments based on rapeseed culture

- **Ensure sufficient oil supply to industrial**



Work on cultural conditions

LUNARIA

- **Sowing** late August, after tillage, with cereal seeder.
Depth 1 - 2 cm.
Density 250 seeds / m² ie **50 - 55 kg/ha** (TKW 20 gr).
First emergence 2 - 3 weeks later
- **Weeding COLZOR TRIO[®] 4 L/ha** at post sowing / pre-emergence,
add **0,5 L/ha of glyphosate** mixed.
In spring, application of **OGIVE[®] 0,4 L/ha + 1 L/ha oil**.
- **Fertilization** estimation of needs = 150 to 200 U of Nitrogen.
Use sulfur fertilizer if possible. P & K = 60 U.
- **Diseases** alternaria, albugo, powdery mildew
Fungicide at bud stage, in april **SUNORG PRO[®] 0,7 L/ha**.
- **Insects** beetles – Treat 1 week after first flowers.
- **Harvest** mid-august directly with combine harvester.
High humidity. Speed 500 à 600 t/mn.
Grain is lightweight : adapt the winds.
Do not fill the hopper, remove the bottom plate.



LUNARIA

History :

August 2008 : from trial in « micro-parcels » to the field (around 2 Ha)

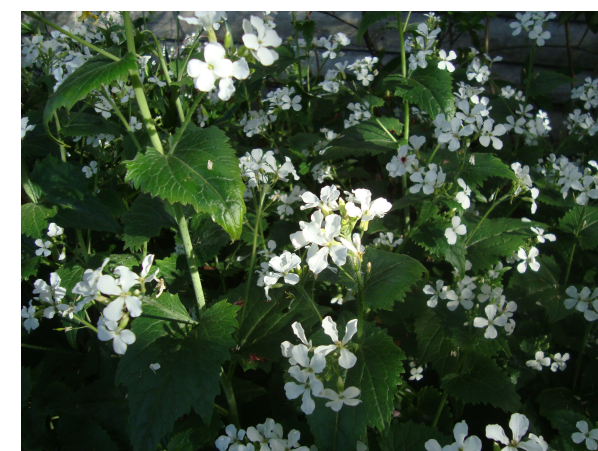
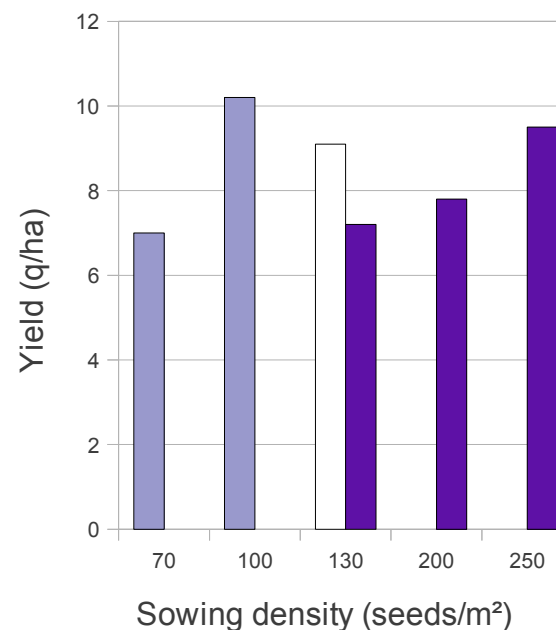
Sowing density (seeds/m ²)	Yield (net) 2009 (q / ha)	Yield 2010 q/ha
70	7,0	
100	10,2	
130	9,1 *	7,2
200		7,8
250		9,5

* Unrepresentative, nb plants germinated < sowing density of 100 s./m²

Explanatory factors :

2010 = very limiting conditions

Limited potential soil (limestone ground)



Optimization

Switching to "field" testing, characterization or confirmation of some constraints

LUNARIA

Problems	Solutions implemented	Results & perspectives
Sowing regularity	<ul style="list-style-type: none"> - test different seeders - «monitor» seeder - increase sowing density - seeds coating 	<ul style="list-style-type: none"> - outweigh regularity problems with nb plants / m² - correct errors with seeder (distribution) - coating penalizing in dry conditions
Weed control	<ul style="list-style-type: none"> - better ground cover - test of herbicides - clean field without weeds, especially cruciferae 	<ul style="list-style-type: none"> - higher sowing density allows better land - use of Round-up at pre-emergence stage. selectivity of Colzor trio, leaf anti-graminaceae catch weeding with Butisan, Lontrel. avoid Cent7. Test Callisto...
Susceptibility to dehiscence High humidity at harvest	<ul style="list-style-type: none"> - desication to homogenize maturity - Swathing ? 	<ul style="list-style-type: none"> - No harvest problem, but high humidity (> 30%)
Grain yield not excellent	<ul style="list-style-type: none"> - test of 5 fields in different pedo-climatic conditions (around 10 Ha) - work on N fertilization - observations of diseases ? 	<ul style="list-style-type: none"> - Better fields - Work in labo



LUNARIA

Work in progress

Grain yield improvement :

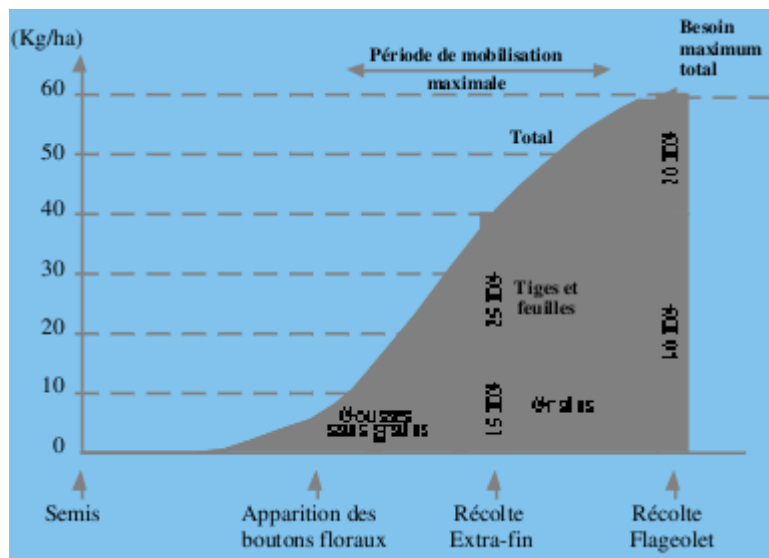
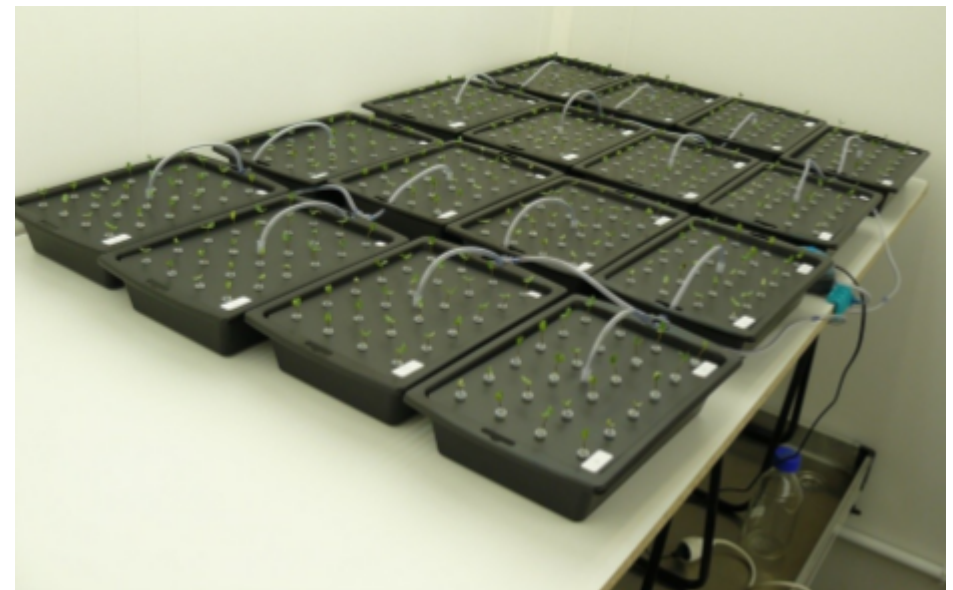
Experiment in laboratory - Hydroponics system

Identification of limiting factors - Bio-elicitation

Mobilization curves on nutrients

Dissociate main (N, P, K, Mg, S) & oligo-elements (B, Mn, Zn)

Evaluate effectiveness of different fertilization to detect, correct any nutritional deficit - Amounts of crop residues



LUNARIA

Work in progress

Oil, erucic and nervonic acid in Lunaria : **Agro-environmental traits influencing their content**

- nitrogen fertilization
- water availability & thermic stress during flowering
- use of some herbicides
- length of filling phase
- abiotic (cold, hydrous) stress
- temperatures
- elongase activity
- photoperiod, solar radiation
- humidity during maturation
- harvest date, plant phytosanitary condition





Exemple of similar work

Omega-3 in linseed : agro-environmental traits

Type:

winter - spring linseed

C 18 :3
(%)

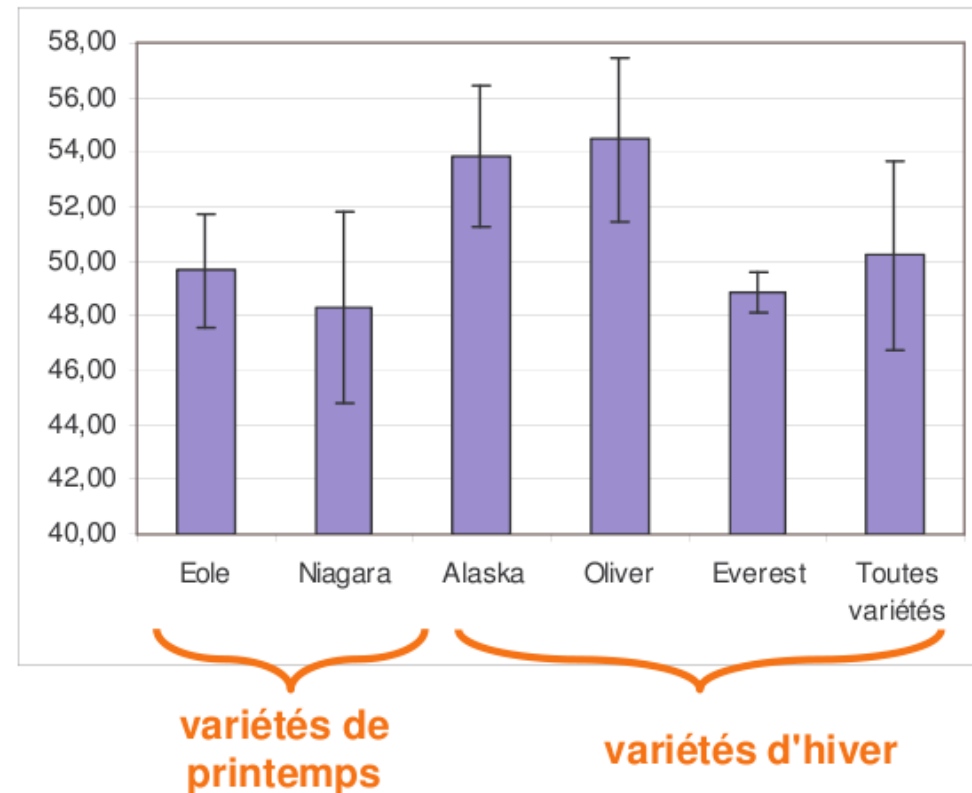


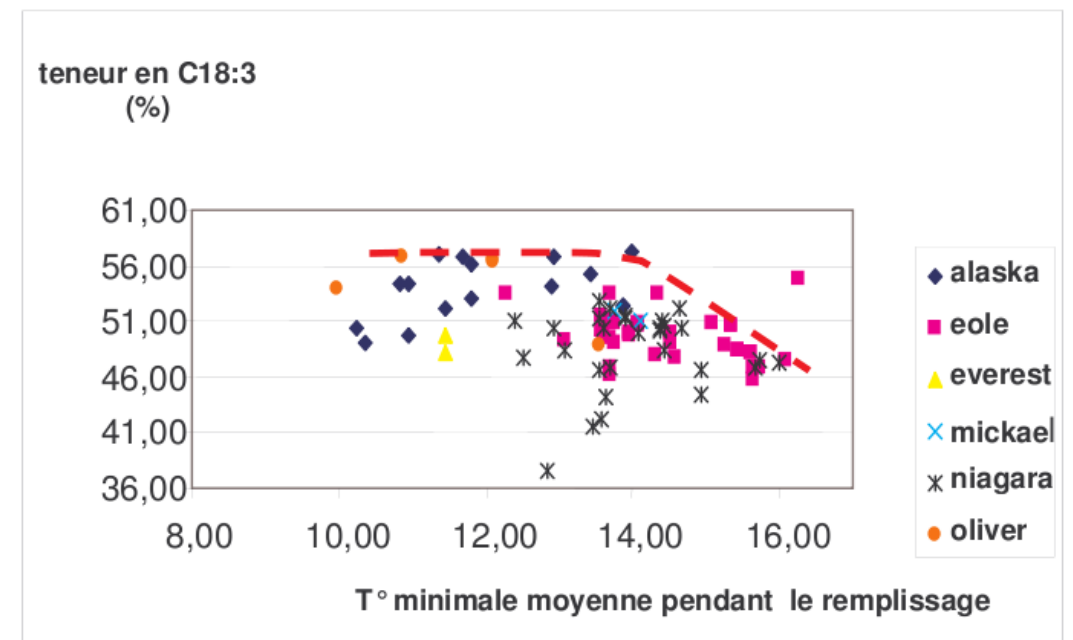
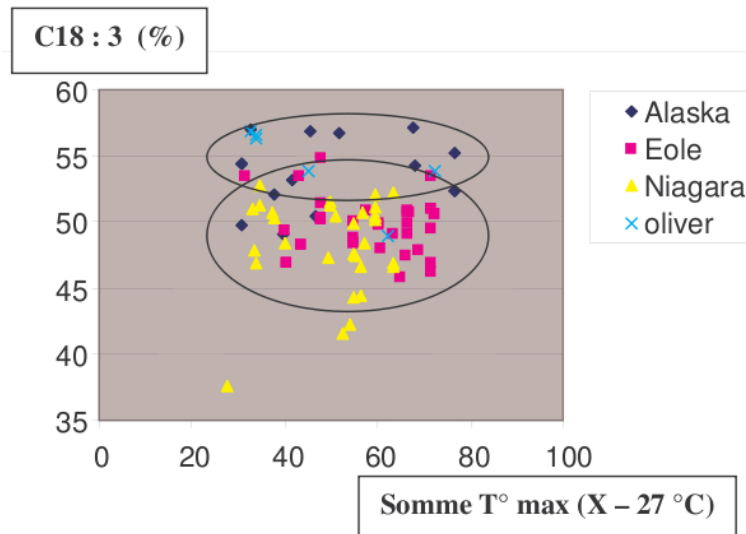
Figure 1 : Teneurs moyennes en C18 :3 et écart-type, pour les différentes variétés

Omega-3 in linseed : agro-environmental traits

Temperature

Potential C 18-3 yield higher when T° during bolls filling are low

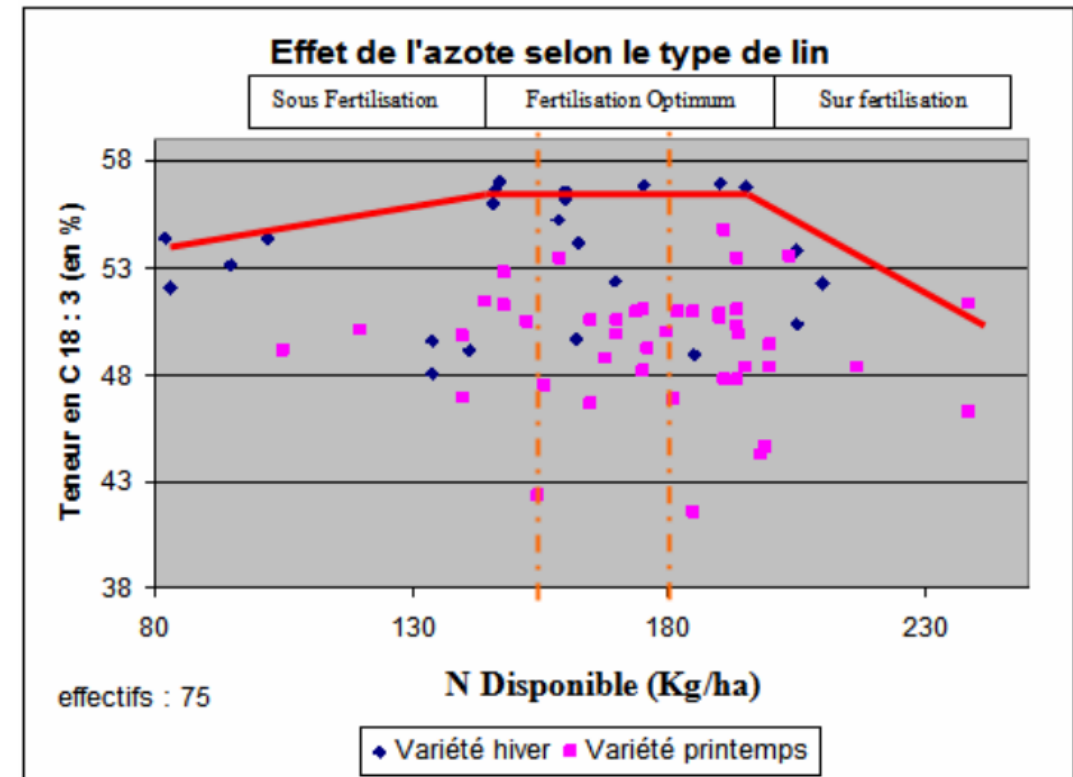
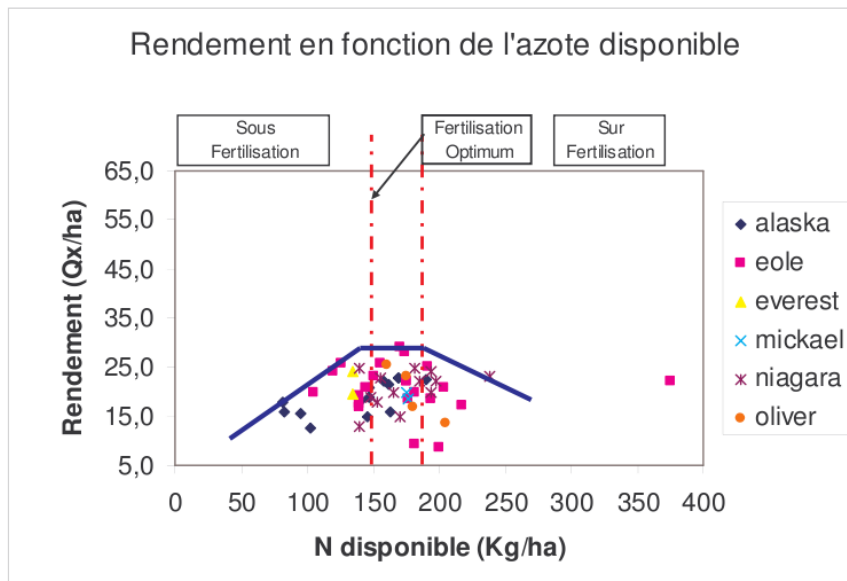
No effect of high T° (thermic stress)



Omega-3 in linseed : agro-environmental traits

Nitrogen fertilization

Best C 18-3 yields for N availability conform to optimum



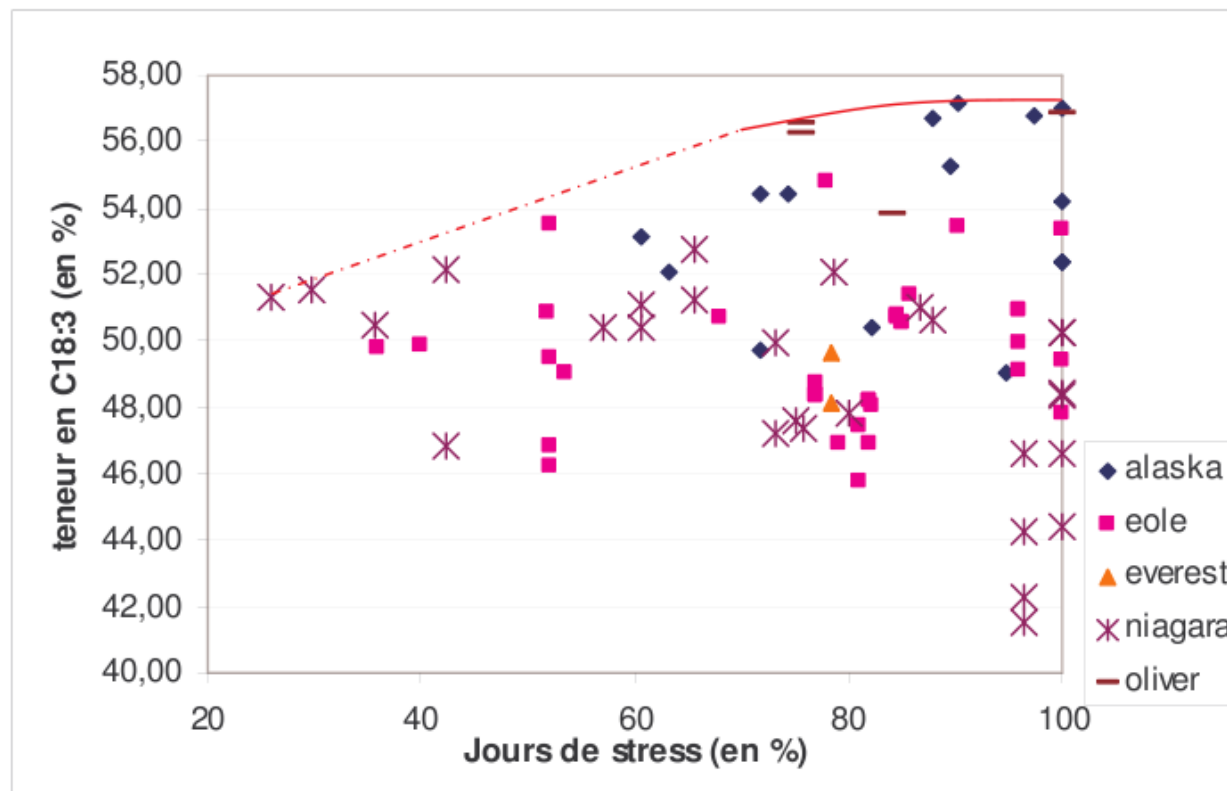
Omega-3 in linseed : agro-environmental traits

Hydrous stress

Water deficit during bolls filling seems to be necessary to get high C 18-3 %

But: significant negative effect on grain yield (flowering time)

Winter linseed: grounds with lower hydrous reserve



Omega-3 in linseed : agro-environmental traits

Hydrous stress and T°

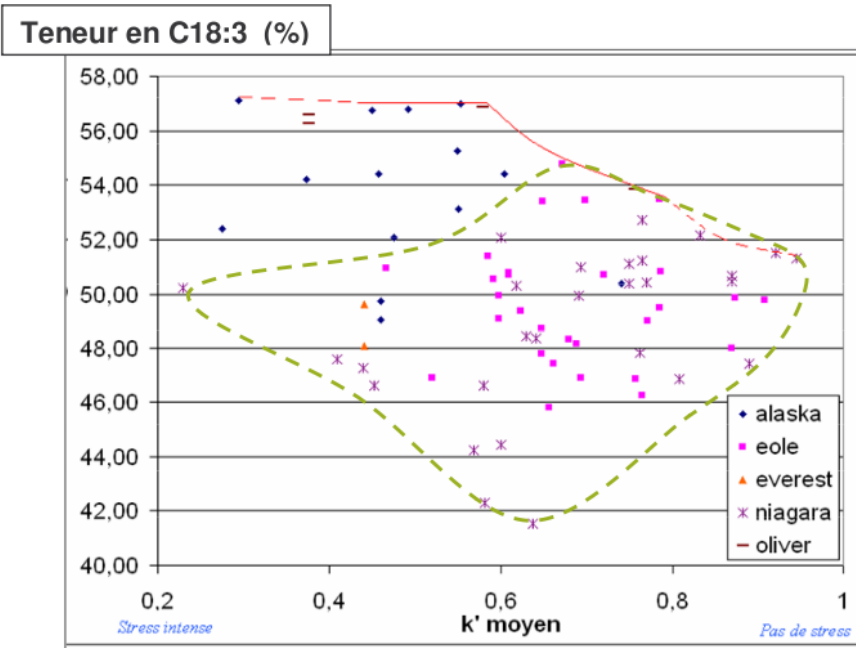
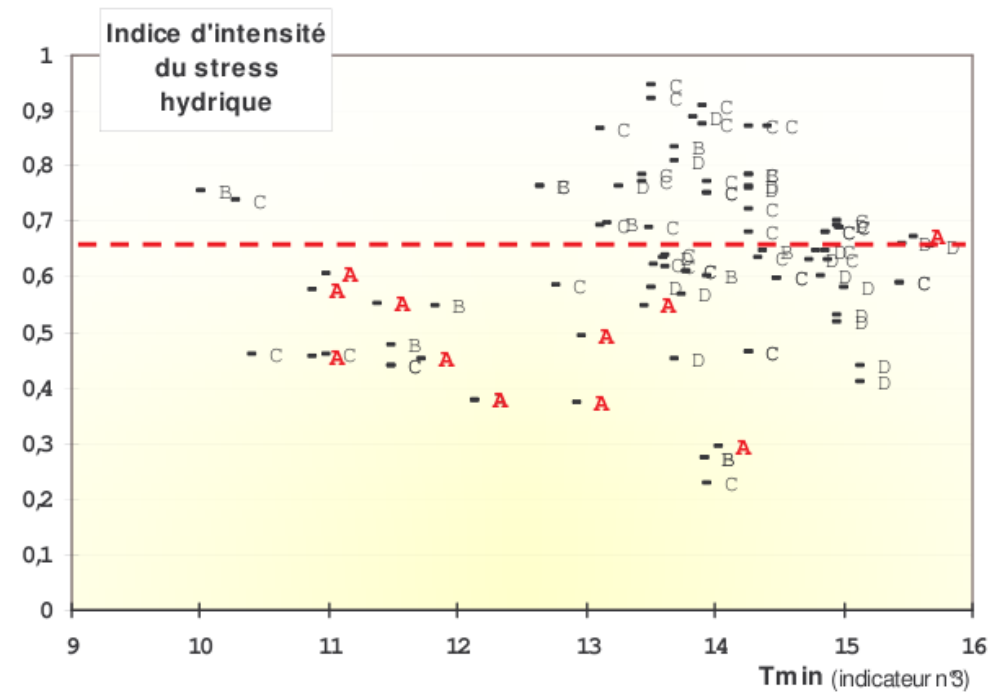


Figure 4 : Teneur en C 18 : 3 en fonction de l'indice d'intensité du stress hydrique durant le remplissage



Omega-3 in linseed : agro-environmental traits

Length of bolls filling

No significant effect observed

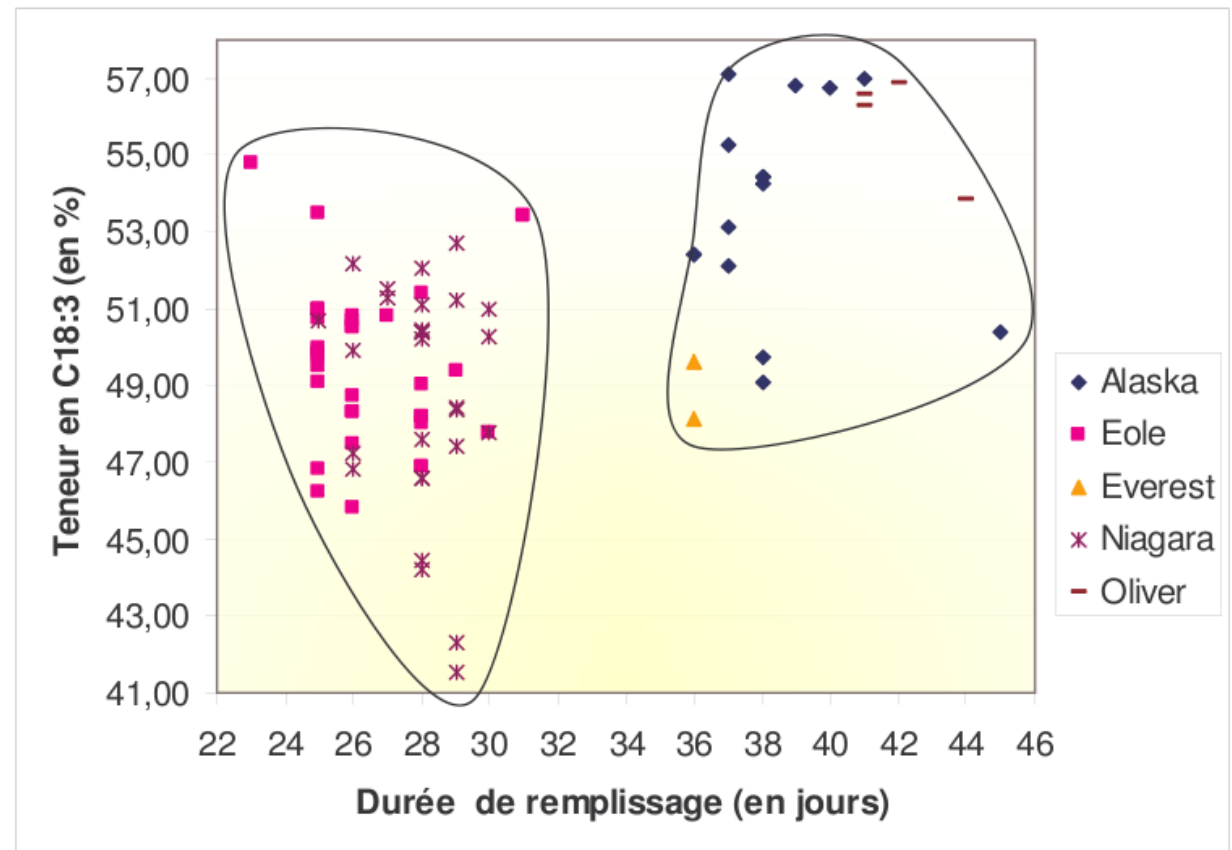


Figure 3 : Teneur en C18 :3 en fonction de la durée de remplissage

LUNARIA

Work in progress

Laboulet

Agronomic aspect : regular yields of 1,5 - 2 t / ha, with 30-35% oil content.

Get Lunaria with a fatty acids profile fitting to the technical, economical and ecological needs of the lubricants industries.

Optimizing Lunaria performances : improving planting density (\approx 55 Kg / ha), in order to answer to the supplying demand, to enter in an economic logic and commercially viability.

Industrial partners

Chemical aspect : develop efficient additives suitable to Lunaria oil

Optimization of oxidation stabilizers used in food industry (fatty acid esters)

Emulgators containing Lunaria having better compatibility with vegetable oils

Work on specific properties of Lunaria oil, allowing to introduce less emulgators and additives in the bio-lubricants

Aim is to achieve the following objectives :

Bio-lubricants having 20% better lubricant properties and 10% less expensive

Formulation aspect : validate the new formulations performances in production and industrial tests applications in cooperation with the end-users



LUNARIA

Conclusion

On technical level, Lunaria presents natural pre-dispositions to the development of these formulations in **lubrication**.

This potential was highlighted during extreme pressure tests where it was more powerful than formulated mineral cutting oils & all vegetable oils used industrially up to date.

The annual variety will be the subject of more breeding and optimization. Lunaria oil will enable to get new generations of **lubricants for the work of metals as for greases based on vegetable oil**.

Economic and environmental stakes are significant and prices of mineral oils are coming close to vegetable oils. Provision of vegetable oils adapted to these applications should facilitate the substitution.

It will allow filling the European criteria of 'Ecolabel' while answering the new lawful criteria, with the respect of the **durable development**.

