

NORTHEAST POTATO TECHNOLOGY FORUM 2005

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Conspectus

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Northeast Potato Technology Forum 2005

Introduction

The Northeast Potato Technology Forum is an annual event that provides potato researchers and extension specialists from Atlantic Canada and Northeastern United States an opportunity to discuss potato research, exchange technical information of benefit to the potato industry, and establish lasting networks. The 2005 event in Fredericton, New Brunswick celebrates the 13th anniversary of the Northeast Potato Technology Forum.

The 2005 program is comprised of 35 oral presentations and 6 posters. The innovative and diverse potato research offered on the program promises to deliver exciting sessions and stimulating discussion. Research topics are all-encompassing: in-field technology, physiology, genetics and breeding, production issues, pests and diseases. Participating authors represent 8 universities and colleges, 5 federal government agencies, 2 provincial governments, and 2 private researchers from 4 Canadian Provinces and 2 American States.

Many thanks are extended to the session chairs, those who provided oral presentations and posters, and to all Forum participants. Our appreciation and thanks go to McCain Foods (Canada) for the publication of this booklet; Potatoes New Brunswick and New Brunswick Department of Agriculture, Fisheries and Aquaculture for technical assistance; and Syngenta for sponsoring the evening reception.

This conspectus booklet contains the abstract or summary of each presentation and poster from the Northeast Potato Technology Forum 2005.

Additional copies of this booklet are available from the New Brunswick Department Agriculture, Fisheries and Aquaculture, Potato Development Centre, 39 Barker Lane, Wicklow, NB E7L 3S4.

Thank you for making the Northeast Potato Technology Forum 2005 a success.

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Soil Amendment Using Composted Pulp Fibre: Effect on Crops of the Second Rotation in Potato Culture in New Brunswick, Canada

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Pulp fibre residue waste from a thermo-mechanical pulping news print process produced in New Brunswick, Canada was composted and the compost used as soil amendment in this second potato rotation. A split application of the compost each of 22.5 tons/ha (1F) and 45 tons/ha (2F), dry weight basis, were used to amend a sandy loam soil in the fall of the year 2001 and 2002, non-amended soil being (0F). Two irrigation protocols of rain-fed supplemented with drip irrigation(I) and rain-fed only (NI) were established.

This soil was previously amended in the fall of the first rotation with one application of 45 ton/ha. (1F) and 90 tons/ha. (2F) of raw pulp fibre residue waste produced from the same process; the soil amendment took place at the end of 1998.

The first crop of this second rotation was Peas followed by Corn, then Potato as the third and final crop of the rotation.

Supplemental irrigation and pulp fibre amendment increased the yield of peas over that of the rain-fed or non amended soil. As for corn and potato crops, yield increased only due to composted pulp fibre amendment of soil under both irrigation protocols.

Total-N in the amended soils of the three year rotation crops at planting and after harvest was higher in amended than non-amended soils in both irrigation systems. Total-N in Pea seeds, corn kernels and potato tubers were also at higher levels when composted pulp fibre was used as soil amendment when compared to non-amended soil in both irrigation systems, these levels of Total-N fall within the normal levels for these crops.

It is believed that the application of this pulp fibre or its compost proved to be a viable agronomic practice to include in potato rotation systems.

Nitrous Oxide Emissions in Potato Production

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Potato production is one of the major agricultural activities in Atlantic Canada, and nitrogen (N) fertilizer inputs to potato are among the highest for agricultural crops. High fertilizer N inputs are required to achieve commercial potato yields, and to achieve the tuber size distribution demanded by the potato processing industry. However, these high fertilizer N inputs also raise concern about emissions of nitrous oxide, a greenhouse gas. Nitrous oxide emissions in humid regions like Atlantic Canada occur primarily through the denitrification process, whereby soil nitrate is converted to nitrous oxide and then to nitrogen gas by soil bacteria under oxygen limited conditions.

Information on nitrous oxide emissions from agricultural production systems is very limited in Atlantic Canada. Research was conducted from 2002 to 2004 to obtain base-line data on nitrous oxide emissions in potato production systems, to determine the effect of the time and rate of fertilizer N application on nitrous oxide emissions, and to determine the effect of landscape position on nitrous oxide emissions.

Nitrous oxide emissions in potato production are generally small in magnitude, ranging from about 0.1 to 4 kg N ha⁻¹. Most of the nitrous oxide emissions occur following significant rainfall events when oxygen becomes limiting under wet soil conditions. Fertilizer N application results in a significant increase in nitrous oxide emissions. There is a trend towards reduced nitrous oxide emissions when fertilizer N is applied as a split application compared to all N applied at planting. Nitrous oxide emissions are substantially higher in poorly drained soils.

It is interesting to note that although soil nitrate concentrations are higher in the potato hill compared to the furrow, denitrification rates are commonly higher in the furrow compared to the potato hill. Also, although nitrous oxide emissions are associated primarily with the denitrification process, denitrification rates are commonly higher in the furrow than in the potato hill whereas the reverse is true for nitrous oxide emissions. These findings emphasize the complex nature of nitrous oxide emissions within agricultural environments. Current research is focussed on improving our understanding of the soil, management and environmental controls on denitrification and nitrous oxide emissions.

Potassium Timing Effects on Potato Yield and Quality

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Our recent research on potash fertilization has shown that potash can have strong positive effects on tuber quality. These effects include decreased susceptibility to internal defects and blackspot bruise. We have also observed improved fry color, increased tuber calcium concentrations, and decreased tuber phenolic concentrations (e.g. tyrosine and other phenolics such as chlorogenic acid). In some experiments where soil test K levels were medium to medium-low, we have also observed yield and tuber size increases. On the negative side, specific gravity has declined by an average of 3.5 points per 112 kg K₂O ha⁻¹ applied at planting. For processing varieties with moderate specific gravity it will therefore be important to supply potash at appropriate rates to provide good internal tuber quality without excessive depression of specific gravity. For fresh market varieties or processing varieties with very high specific gravity, it will be important to supply potash at sufficient rates to maximize internal quality and provide moderate specific gravity. These results increasingly point toward potash fertility programs that are specific to soil test results for a field, intended market, and an individual variety's characteristics. Prior to 2002, our research focused on at-planting potash treatments. Growers need flexible management programs and they expressed an interest in the effectiveness of potash applications applied after planting. This paper will discuss results from a three-year study designed to evaluate the effectiveness of fertility programs in which a portion of the potash was removed from the at-planting application and applied at a later date.

An experiment using Russet Burbank was conducted during 2002-2004 at Aroostook Research Farm in Presque Isle, ME. Treatments consisting of 0, 112, 224, or 336 kg K₂O ha⁻¹ applied at-planting were used to establish the rate response characteristics for the experimental sites. The at-planting potash treatments were compared to four treatments in which a portion of the potash was applied at planting and the remainder was applied after planting, either before cultivation or at last hilling. These treatments were as follows: 1) 112 kg K₂O ha⁻¹ at planting followed by 224 kg K₂O ha⁻¹ before cultivation; 2) 112 kg K₂O ha⁻¹ at planting followed by 224 kg K₂O ha⁻¹ at last hilling; 3) 224 kg K₂O ha⁻¹ at planting followed by 112 kg K₂O ha⁻¹ before cultivation; and 4) 224 kg K₂O ha⁻¹ at planting followed by 112 kg K₂O ha⁻¹ at last hilling. The potash source was muriate of potash (KCl) for all treatments. Response to the fertility program was measured under irrigated and non-irrigated conditions. We measured foliage nutrient concentrations, total nutrient uptake, yield, and tuber quality (e.g. size distribution, internal defects, specific gravity, bruise susceptibility, fry color, and

tuber sugars). These treatments and response variables allowed us to determine if the majority of the potash could be removed from the fertilizer band and also whether the delayed potash needed to be applied soon after planting or if it could be delayed until hilling.

The potash fertilizer rate responses in these studies matched those described above for our earlier research. Foliage analysis showed that there was a measurable lag in K uptake from the delayed potash treatments, but that by the end of the growing season there was no significant difference in uptake between the at-planting and delayed-potash treatments. The results also generally demonstrate that the delayed potash treatments had comparable yield and quality to the at-planting treatments. We conclude that up to 2/3 of the recommended potash can be removed from the fertilizer band and applied at a later date without penalty. While we did not observe any significant yield/quality benefits or penalties from delaying some of the potash in these trials, the flexibility in timing and placement of potash could be useful to growers as they manage the logistics of a busy planting season, variable soil test K among fields, and varieties with different optimum potash requirements.

Potential Indicators of Soil Quality of Potato Fields in the Maritimes

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Although conventional agriculture has increased yield in the short term, the intensification of agricultural inorganic input, including fertilizers and pesticides, may be detrimental to the sustainability of agriculture and the environment. As an alternative, organic agriculture emphasizes the importance of soil biological activity as a means of growing higher yielding, pest free products. This begs the question, are organic soils actually healthier than conventional soils? Soil quality, defined by Doran and Parkin in 1994, is a concept that is used to evaluate soils fitness for function in managed systems. There is a need for a soil quality index that would use a combination of biological, physical, and chemical indicators to qualitatively evaluate agroecosystems under differing management systems. Using eleven standard indicators of soil quality, conventionally and organically managed potato fields in New Brunswick, Prince Edward Island, and Nova Scotia were compared over the 2004 growing season. Although there were very few studies that served as soil quality indicator baselines in the Maritimes, benchmarks were gathered from studies of similar climates and management practices. Mites (Acari), collembola, nematodes, testate amoebae, microbial biomass, cellulase assays, and ATP extraction served as bioindicators, while bulk density, soil moisture content, light fractionation, and pH served as the physical and chemical indicators. Preliminary analysis has shown no statistically significant difference between conventionally and organically managed potato fields. Further sampling in a no-tillage organic potato field resulted in greater diversity of soil biota. This suggests that the effects of chemicals on the state of nutrient cycling through decomposition may be less destructive than the physical disturbance of tillage. Further studies using bioindicators of soil quality are needed to increase the baseline values used in soil quality indexing in the Maritimes, and for further investigation of tillage and no-till practices.

Influence of Mulch and Water Application on Field-scale Soil Water Patterns in Maine Potato Production

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Past research indicates that field-scale soil water patterns are stable in time and have a strong bearing on water application requirements and yield. However, little is known about how to manage this variability in Maine potato production. Soil water content was measured biweekly at 30 m intervals using a portable hammer-driven time domain reflectometry system over six field-scale transects in potato (*Solanum tuberosum* L.). Transects were split into mulch and no-mulch, irrigated and un-irrigated treatments in 2004 and yield was measured using hand sampling. Supplemental irrigation was applied once in the amount of 2.5 cm. Trends in yield vs. soil water content suggested yield increased with soil water over the drier parts of the field but declined at higher water contents under all treatment scenarios. Light straw mulch (around 45% ground cover) application had no discernible effect on yield or relative water status. The 2004 growing season had adequate moisture from rainfall. Under these conditions, only the drier areas of a field appear to benefit from the type of heavy, infrequent supplemental irrigation regime used in this study.

Evaluations of Nutrient Management Fertilization with the Potato Varieties Shepody and Russet Burbank under Dryland and Irrigated Regimes

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Introduction

The energy for our everyday human activities comes from the sun. Green plants capture the energy from the sun and convert it to sugar and starch. Seventeen essential nutrients are required for plant growth. If an essential nutrient is absent, a plant cannot complete its life cycle. Macro nutrients such as nitrogen, phosphorous and potassium are required in large quantities (macronutrients); whereas, micro nutrients such as copper, nickel and zinc may be required only in very small quantities and are generally available from the soil..

When the first settlers came to PEI, it was covered with conifers and deciduous trees. Much of the land was cleared and planted to crops. For the first few years, good crops were obtained, as the supply of available nutrients in the soil was removed and not replaced. The soil became depleted and crop yields declined. It is important to recognize that every product that leaves the farm (vegetables, livestock, potatoes, grain) is a drain on the soil.

The introduction of concentrated chemical fertilizers became widespread in PEI in the 1950s and facilitated the provision of the three macro nutrients nitrogen, phosphorous and potassium (potash). Many farmers and gardeners like to be “kind” to their land and crops; hence some apply generous quantities of fertilizer. If the crop does not use all of the nitrogen fertilizer, the nitrate component can leach into the ground water. A number of wells in PEI have higher nitrate concentrations than the internationally accepted tolerance (10 ppm). Phosphorous and potassium are much less mobile but surface run-off of phosphorous from fields by erosion can lead to algal blooms in ponds and streams. There has been increasing concern regarding eutrophication in rivers and streams.

Around the world, there are increasing regulations to restrict over-fertilization of farmland. In some European countries, farmers may not be able to spread manure on their land if soil tests reveal that high concentrations of nutrients are already present in the land. Use of concentrated fertilizers is subject to audits. On PEI, extension personnel in government and private industries are working with some growers to explain the concepts of nutrient management and to conduct trials under PEI conditions. Field scale research projects and demonstrations are being conducted to document the yield and quality

responses to different fertility regimes. Once the nutrient content in manure and the nitrogen contribution from legume crops is determined there would be fewer requirements for chemical fertilizer. Some of the growers who are participating in nutrient management trials recognize that financial returns may still be very acceptable with reduced fertilizer applications; and that environmental risks have also been decreased. Nutrient management will be an important issue for everyone on PEI for financial and sustainable practices.

Objectives

As farmers have generally decided to err on higher than required amounts of fertilizer applications, a considerable amount of local data on the yield and quality of potatoes from different fertilizer treatments will be required to gain their acceptance for nutrient management rates. The objective of this study was to assess financial returns, under the industry's processing contract, for potatoes fertilized by traditional and nutrient management rates. The crop response under irrigated and non-irrigated conditions was assessed

In an effort to build a "bank account" type of approach, soil samples were taken before the potatoes were planted and after the potatoes were harvested to determine residual amounts of P, K, Ca and Mg. The generation of such information will help to determine changes in nutrient concentrations in soil when potato crops are removed from fertilized and non-fertilized land. It will also help in determining the optimum concentrations of P and K in soil tests from PEI for optimum fertilization of Shepody and Russet Burbank.

Methods and materials

Nutrient management trials were conducted at Cavendish Farms, New Annan, and Prince Edward Island in 2003 and 2004 under irrigated and non-irrigated conditions. The site of the replicated field trials was in a field where three year rotations had been practiced for many years (potatoes, barley -under seeded, mixed clover grass hay). The field was treated with glyphosate herbicide in the fall and "Res-Tilled" in the spring of each year. Soil samples were collected and submitted to the Charlottetown Soil and Feed test Laboratory, for complete soil test evaluations. Nutrient Management recommendations were made by personnel from the PEI Dept. of Agriculture. The "traditional" farmer applications (Farmer) were determined from discussions with technical and sales personnel from the fertilizer industry. All fertilizer mixes were banded at planting. In the 2003 trials, no limestone was applied. In 2004, dolomitic limestone was applied to all treatments. Soil samples were collected from all replications after the potato harvest to verify residual concentrations of P, K, Ca and Mg.

The two varieties used in the trials were Shepody and Russet Burbank. Seed pieces were planted at 10" spacing for Shepody and 18" for Russet Burbank. Four replications were planted for each of the fertility treatments. Identical trials were conducted in the same field

under an irrigated regime (Center pivot system) and non irrigated conditions. In 2003, a total of 1.5 inches of water was applied in three applications. In 2004 a total of 3.25 inches was applied in four applications. A decision to irrigate was based on readings from a Hydro-sense meter and Irrrometer soil probes. Soil tests, taken in the spring of 2003 - 2004 revealed the phosphorous (426ppm – 531ppm) and potassium (192ppm – 232ppm). Levels were in the high range. In 2003 and 2004 the three treatments were, check plots (no fertilizer), nutrient management, and farmer. In 2004, due to a low pH and relatively low magnesium concentrations, Dolomitic limestone was applied, at a rate of one ton per acre to all plots by spreading over the hills and hilling in.

All potato samples were graded to contract specifications for the Cavendish Farms processing contract. Tubers are sorted into one of three categories <2" diameter, > 2" but less than ten ounces and > ten ounces. The specific gravity in each replication was determined from a sample of 25 tubers. These tubers were also cut to determine the weight of tubers with internal defects i.e. hollow heart, brown center, stem end discoloration, vascular ring discoloration. The data from the plots was used to calculate the financial returns per acre, based on a current winter processing contract. Fry colors were rated for each replication.

In 2003, the nutrient management rate (pounds per acre of nutrients) was lower for N, P and K for both varieties. In 2004, the nutrient management rate was identical for nitrogen applications for Shepody and also for Russet Burbank, but lower for P and K (table 2). The fertilizer mixes were made from ammonium nitrate, di-ammonium phosphate and potassium chloride.

Results and discussion

The nutrient content of the soil before fertilization and after removal of the crop is summarized in table 1. In both 2003 and 2004, the concentration of Phosphorus and Potassium dropped after crop removal from non-fertilized plots. There was a slight drop in the Potassium in nutrient management plots, and Phosphate stayed in the same range. For the farmer, mix there was a trend to increased concentrations of Phosphorus.

The yield and financial value of the crops in each treatment is shown in (table 3).

The addition of fertilizer increased the yield of both varieties. Irrigation increased the biological yield of tubers in 2003 and 2004. However in addition to the increased biological yield, slightly darker fry colors were observed in irrigated and fertilized trials (delayed maturity), Specific gravity was slightly less in irrigated trials. Hollow Heart was also more extensive in irrigated trials, especially in Shepody in 2004 trials. The gain made in the increased yield of tubers (Shepody variety) was lost due to dockage for the increased Hollow heart and sunburn. Even though non-fertilized Shepody tubers were smaller than those from

fertilized plots, extensive Hollow Heart was observed in the non-fertilized and irrigated Shepody tubers. The increased yields in irrigated Russet Burbank potatoes resulted in substantial financial returns compared to the non-irrigated Russet Burbank in 2004.

In 2003, the higher fertilizer rates in Russet Burbank gave improved yields compared to the nutrient management rate in both the irrigated and non-irrigated plots. In 2004, yields and financial returns were similar for the “nutrient management” and “farmer” rates for irrigated Russet Burbank. In the non-irrigated Russet Burbank, the “farmer rate” gave higher yields. In 2003, the financial returns in the Shepody variety were similar for both fertility rates in irrigated and non- irrigated conditions. In 2004, although substantial yield increases were noted in both fertility rates when irrigated, the dockage due to Hollow Heart and Sunburn resulted in similar financial returns from the irrigated and non-irrigated Shepody plots.

A consistent observation was for reduced specific gravity under the higher fertility applications.

The research program will be continued to build a data bank.

Table 1

		Fertilizer Trial Soil Testing Non Irrigated				
		pH	Phosphate (ppm)	Potash (ppm)	Calcium (ppm)	Magnesium (ppm)
2003	<u>Spring</u>	5.8	426	192	794	104
2003	<u>Fall</u>					
	<u>0 – 0 – 0</u>	5.8	386	151	941	106
	<u>Nut-Man</u>	5.6	414	157	860	94
	<u>Farmer mix</u>	5.5	710	219	877	106
2004	<u>Spring</u>	5.4	532	232	462	56
2004	<u>Fall Shepody</u>					
	<u>0 – 0 – 0</u>	5.9	482	171	624	175
	<u>Nut-Man</u>	5.4	579	186	570	139
	<u>Farmer mix</u>	5.6	589	211	608	174
2004	<u>Fall Russet Burbank</u>					
	<u>0 – 0 – 0</u>	5.7	465	154	575	142
	<u>Nut-Man</u>	5.6	535	154	556	163
	<u>Farmer mix</u>	5.2	583	200	481	119

Table 2

Fertilizer Mix

	2003			2004		
<u>Shepody</u>						
Nut.Man.	126 N	90 P	122 K	160 N	135 P	135 K
Farmers	130 N	200 P	200 K	160 N	170 P	220 K+Kmg
<u>Russet Burbank</u>						
Nut.Man.	144 N	122 P	122 P	180 N	135 P	135 P
Farmers	180 N	200 P	200 P	180 N	200 P	220 K+Kmg

Table 3

	2003				2004			
	Net	%	%	Total	Net	%	%	Total
	<u>Weight</u>	<u>Hollo</u>	<u>Sunburn</u>	<u>Payable</u>	<u>Weight</u>	<u>Hollow</u>	<u>Sunburn</u>	<u>Payable</u>
	per	w		per	per			per
	acre			acre	acre			acre
<u>Shepody Irrigated</u>								
0 - 0 - 0	24061	7	2	\$1,547	24,108	39	4	\$986
Nut-Man	32277	2	8	\$2,185	35,228	17	11	\$2,122
Farmer	35424	21	5	\$2,065	34,401	15	15	\$2,066
<u>Shepody Non-Irrigated</u>								
0 - 0 - 0	21023	0	0	\$1,402	12,851	9	0	\$728
Nut-Man	29937	3	4	\$2,013	28,195	3	1	\$2,073
Farmer	31559	2	8	\$2,173	28,802	2	6	\$2,154
<u>Russet Burbank Irrigated</u>								
0 - 0 - 0	20666	4	0	\$1,163	18,246	8	1	\$944
Nut-Man	30774	13	1	\$1,658	32,930	4	3	\$2,247
Farmer	37832	10	2	\$2,545	33,169	6	2	\$2,312
<u>Russet Burbank Non-Irr.</u>								
0 - 0 - 0	16566	6	0	\$761	10,494	0	1	\$396
Nut-Man	28139	6	0	\$1,780	23,963	4	3	\$1,333
Farmer	35608	10	5	\$2,121	29,056	7	1	\$1,869

Feb.21

An Economic Evaluation of Raised Bed and Use of Green-Sprouted Seed Tubers in the Maine Potato Cropping System

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Management practices that accelerate crop development and allow earlier plant establishment would be beneficial in short-season potato (*Solanum tuberosum* L.) production areas. A two year study in northern Maine was conducted to determine the yield and quality effects on the cultivar Russet Burbank attributable to tillage system and seed sprouting treatments. In this analysis two tillage systems, consisting of fall raised bed (RB), and spring chisel plow (CH) in combination with non-sprouted seed and green-sprouted seed, are compared. For the purpose of the economic analysis fall raised beds with green sprouted seed is compared to the spring chisel plow tillage system and non-sprouted strategy as this represents the industry standard. To determine the potential economic efficacy of fall raised beds and green sprouted tubers partial budgeting analysis was used. Partial budgeting is employed to evaluate small changes to a production system with respect to system profitability. Under the cost and yield assumptions used, a five percent increase in yields is necessary to justify the adoption of green-sprouted tuber in raised beds. The key costs parameters are sufficient space to produce the green sprouted seed and racking system to handle the tubers.

Non-destructive Estimation of Potato Leaf Chlorophyll and Nitrogen Contents from Spectral Measurements

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Optimizing N fertilization in potato production by in-season measurements of potato N status may improve fertilizer N use efficiency. Hyperspectral leaf reflectance and transmittance measurements may be used to assess potato N status by estimating leaf chlorophyll or N contents. This study evaluated the ability of the inverted PROSPECT radiative transfer model to predict leaf chlorophyll and N (as protein) contents. Trials were conducted with 'Russet Burbank' and 'Shepody' potato cultivars under different N fertility rates (0-300 kg N ha⁻¹) in 2001 and 2002. Leaf reflectance and transmittance, leaf chlorophyll content and leaf protein content were measured. Leaf chlorophyll and protein content were well correlated, but the relationship was strongly dependent on sampling date. Chlorophyll content was predicted with reasonable accuracy by the model, particularly in the 2002. The low estimation accuracy in 2001 was probably related to sample variability induced by prolonged drought conditions. Protein content could not be predicted with any degree of accuracy by the model. The relative success of the PROSPECT model to predict chlorophyll content, and the good correlation between leaf chlorophyll and leaf N, suggests that it might be used as a component of a more complex leaf-canopy reflectance model to estimate chlorophyll content from reflectance spectra at the canopy level.

Starch Phosphorylase Enzymatic Activity And Dry Matter Accumulation in Developing Potato Tubers

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Introduction: Starch phosphorylase (STP) catalyzes a reaction that can degrade as well as synthesize starch if tested under in vitro conditions. However, the role of STP for starch synthesis in plant cells is poorly understood, while another enzyme, AGP-glucose pyrophosphorylase (AGPase), is known to be an enzyme essential for starch synthesis and dry matter accumulation. To study the role of STP in dry matter accumulation in developing potato tubers, we conducted a comparison of the diurnal change pattern of STP activity with that of plant photosynthesis rate and that of AGPase activity.

Materials and Methods: Greenhouse plants of two potato cultivars ('Shepody', a tetraploid; and '11379-03', a diploid) at the rapid tuber-bulking phase were used to measure enzyme activity, photosynthesis rate, and dry matter content. Sampling of leaves and tubers was conducted at 5 AM, 9 AM, noon, and 9 PM.

Results and Discussion: Dry matter of tubers in both cultivars increased during the day, about 1% for Shepody and 1.5% for 11379-03 from 5:00 AM to 9:00 PM, respectively. STP enzymatic activity had a similar diurnal pattern to that of AGPase in both leaves and tubers of the two cultivars, of which maximum total activity was recorded at noon. However, the activity of STP was much higher than that of AGPase. STP activity in tubers was also found to be correlated with the photosynthetic activity in leaves. These results suggest that STP does play a positive role in dry matter accumulation in developing potato tubers.

The Full Spectrum: Research on Potatoes with Coloured Flesh at PRC

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The adapted potato germplasm resource at AAFC contains a wide array of materials that offer research and development opportunities. Diploid germplasm developed initially to study the inheritance of skin and flesh colour in potatoes and the association with eye depth now is providing the base for new research. With growing scientific and public interest in the merits of anti-oxidants present in fruits and vegetables, attention has returned to potatoes with purple and red pigmented flesh. A research project to improve and identify superior selections has been underway at Fredericton since 2003.

Extracts from these potatoes are rich in flavonoids, predominantly anthocyanins, which have antioxidant activity. The reported benefits of antioxidants make them an important component of a healthy diet. The germplasm collection contains materials with pigmented flesh in a range of patterns ranging from coloured rings through star bursts to full pigmentation. Measured levels of anthocyanins from these materials have exceeded those found in commercial cultivars by three to four times and some will also produce attractively coloured chips.

This paper will provide an overview of multi- disciplinary research at the Potato Research Centre describing progress and future directions.

Cluster Analysis of Potato Heritage Varieties Based on DNA Fingerprints

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Heritage potato cultivars (often called heritage potato varieties) were collected and grown by gardeners, farmers, and scientists in Canada before the establishment of a formal cultivar registration system in 1923. Canadian seedling and clonal selections, and introductions from other countries, continue to be valued and maintained today outside the certified seed system for the production of registered cultivars. Heritage potato cultivars may also be useful germplasm for potato breeding.

Old cultivars are often known by different names for a number of reasons including having been collected from different regions of the country where they were given a "local" name. Hence today there is some confusion regarding the identity of these cultivars. Heritage cultivars can only be properly managed and utilized if they are correctly identified in germplasm collections.

At the Potato Research Centre new DNA protocols have been developed to distinguish genetically close potato cultivars. In the present study, our protocols were based on polyacrylamide gel electrophoresis of amplified DNA fragments containing simple sequence repeats and other polymorphic DNA sequences. These protocols have been used to analyze heritage cultivars, and a cluster analysis of the cultivars based on the resulting DNA fingerprints has been conducted.

The DNA samples of cultivars for the analysis were obtained from three sources: 1) the Potato Gene Resources Repository, Agriculture and Agri-Food Canada, Fredericton; 2) Canadian Potato Variety Repository, operated by NB Dept of Agriculture, Fisheries and Aquaculture, Fredericton; and 3) Seeds of Diversity Canada (Mr. Garrett H. Pittenger), Ontario.

In our study DNA fingerprints were found to be useful to sort out confusion caused by mis-

named heritage cultivars. The results indicated that some heritage potato cultivars have identical fingerprints. This phenomenon is particularly frequent within the blue flesh cultivars and the fingerling cultivars.

The information generated from this study will contribute to the effectiveness of conservation, evaluation, and utilization of heritage potato cultivars.

Inheritance Mode and Genetic Mapping of Tuber Eye Depth in Cultivated Diploid Potatoes

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Tuber eye depth is an important trait for processing quality and appearance. During the past century several hypotheses, sometimes contradicting each other, have been proposed on the inheritance of eye depth. Considering that most of these earlier investigations were carried out with relatively small populations of tetraploid *S. tuberosum* progenies, such confusion is not surprising. This study was conducted with cultivated diploids because:

1. During the last several decades populations of well-adapted, fertile cultivated diploid hybrids (*S. phureja* x *S. tuberosum* haploids and *S. stenotomum* and *S. tuberosum* haploids) which produce medium and large tubers under New Brunswick field conditions have been developed at the AAFC Potato Research Centre. This material is very well suited for genetic studies of tuber traits.
2. Genetic studies can be done much easier with diploids ($2n=2x=24$) than with tetraploid ($2n=2x=48$) *S. tuberosum*. The disomic inheritance pattern of the diploids allows for much simpler ratios than are possible with the tetrasomic pattern of the tetraploids.
3. As result of (1) and (2) above the AAFC diploid populations have become a platform technology for further genetic research on potatoes.

Most of our work was done with diploid family 12601. The parents of this family were 11379-03 (shallow eyes, long tubers) and 08675-21 (deep eyes, round tubers). We found that a major gene controls eye depth. The gene symbol *Eyd* is proposed. Deep eyes (*Eyd*-) are dominant over shallow eyes (*eyd/eyd*). Deep eyes tended to be associated with round tubers and shallow eyes segregated jointly with long tubers. Similar results were obtained with another family (Family 12586; data not shown).

In a previous study it was found that a major locus controlling tuber shape (*Ro/ro*) is located on chromosome 10; *Ro*- produces round and *ro/ro* long tubers (De Jong and Burns, 1993). In the present evaluation of families 12601 and 12586 with molecular markers, including SSRs and AFLPs we found that the *Eyd/eyd* locus is located on chromosome 10 and closely linked to the *Ro/ro* locus at a distance of about 4 Centimorgans.

Reference.

De Jong, H. and V.J. Burns. 1993. Inheritance of tuber shape in cultivated diploid potatoes.

Amer. Potato J. 70: 267-283.

The Accelerated Release of Breeding Selections from the Potato Research Centre: A Review

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In the mid 1990s Agriculture and Agri-Food Canada reviewed the potato cultivar release procedure for the breeding program at the Potato Research Centre. The impact of cultivars released from the breeding program had been limited, with two major exceptions, to a regional basis. Also, the time to bring new cultivars to market was a minimum of 12 years, and frequently more, by a trial system that generated sufficient data for cultivar registration which was then followed by block or grower trials. The cultivar then had to be established in the seed certification system before it was available for commercial production. At the same time, some sectors of the industry were asking for earlier involvement in cultivar evaluation.

Agriculture and Agri-Food Canada required a new release system to lead to an improved rate of adoption of AAFC cultivars, to reduce the time to commercialization, to be sensitive to market forces and to use a fair and open process.

A two stage “accelerated release” procedure was developed to meet these requirements. In stage one, industry is offered new selections for two years of non-exclusive field and post-harvest testing. Following the completion of the non-exclusive testing, companies interested in further testing are invited to submit cash bids with the highest bidder advancing to stage two and procuring exclusive testing for a further three years. At the end of this testing, or sooner at the request of the company, stage two also allows the company to negotiate with AAFC a six-year, renewable licence to commercialize the selection. This procedure was adopted in 1998 for French fry selections and in 2003 extended to include chip and fresh market selections. The breeding program is committed to release selections each year, and promotes the selections and the release procedure at field days, an open house and on the AAFC website.

Sixty two selections have been offered to industry since 1998, including a transition group of 23 advanced selections that were in the trial system when the “accelerated release” procedure was adopted. Data will be presented on industry involvement in the non-exclusive and exclusive stages of evaluation, and participation by sector and geographic region.

Identification of Proteins Involved in After-Cooking Darkening in Potato

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After-cooking darkening (ACD) is an undesirable trait in potatoes worldwide. It occurs upon exposure of potatoes to air after cooking. It is widely accepted that the cause of ACD is the formation of an iron-chlorogenic acid complex which oxidizes upon cooling to form a dark color. ACD is controlled genetically and influenced by environmental factors. It is believed that there are genes expressed and hence proteins that are involved in controlling the severity of ACD. Proteomics, the study of the protein complement of the genome, shows all the proteins expressed under certain conditions at a certain time. Proteomics is being used in this study to find proteins that are expressed or repressed in potatoes that are susceptible or resistant to ACD. Tubers of diploid varieties with varying degrees of susceptibility to ACD, as well as tubers of tetraploid varieties grown in different locations are being used as comparisons. A number of proteins have been identified by LC-MS/MS followed by online database searching. Further research is expected to show which proteins are necessary for ACD.

Potato Tuber Color Genes And Their Use In Variety Development

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Over the past several years our research group has identified genes associated with red and purple skin color, as well as yellow tuber flesh. In particular, we have identified an allele of dihydroflavonol 4-reductase (dfr) associated with the production of red anthocyanin pigments, an allele of flavonoid 3', 5'-hydroxylase associated with production of purple anthocyanins, an allele of an R2R3 myb domain gene associated with accumulation of red or purple anthocyanin pigments in tuber skin, and an allele of beta carotene hydroxylase associated with increased accumulation of yellow carotenoids in tuber flesh. Simple PCR assays to track the alleles associated with red skin color and yellow tuber flesh have been developed. For the red allele of dfr, we have also developed an assay to measure dosage of the functional allele (nulliplex, simplex, duplex, triplex, quadruplex) relative to other alleles. These assays allow us to more rationally manipulate color genes in our conventional potato breeding program. Should transgenic potatoes ever be accepted in the marketplace, functional alleles of color genes will also provide opportunities for altering color of existing varieties, e.g., converting a white variety into a red-skinned variant, or a red-fleshed cultivar into a purple-fleshed one.

Gene Expression Studies on Genetic Control of After-Cooking Darkening in Diploid Potatoes

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Potato (*Solanum tuberosum* L.) is one of the major staple food crops. It ranks fourth in the worldwide production after rice (*Oriza sativa*), wheat (*Triticum aestivum*) and maize (*Zea mays*). The quality of the potato tuber is one of the most important characteristics in determining the quality of the crop. Tuber appearances, such as flesh color, skin color, and discoloration after cooking can influence consumer preference. Processed or cooked potatoes often darken when exposed to air. This phenomenon, which is an unattractive trait of cultivated potatoes worldwide, is known

as after-cooking darkening (ACD). It is generally understood that ACD is caused by a non-enzymatic oxidation reaction in which the ferrous-chlorogenic acid complex formed in the tubers during cooking oxidizes to form a bluish-gray colored ferri-dichlorogenic acid when exposed to air. Many investigations have found a relationship of the degree of the darkness with some plant metabolites besides chlorogenic acid, such as citric acid, ascorbic acid and iron. Combination of genetic factors and environmental conditions determines the overall content of these chemicals, resulting in the differences in the susceptibility of ACD in potatoes. The environmental effects directly impact the farm practices, whereas understanding the genetic control mechanisms would lead to the new cultivar development.

In this study, the differential gene expressions of four candidate genes were tested. They are two genes responsible for the chlorogenic acid biosynthesis (genes encoding *cinnamate 4 hydroxylase* and *hydroxycinnamoyl CoA quinate transferase*), one gene responsible for citric acid metabolism (gene encoding *citrate synthase*) and one gene for iron capturing protein (gene encoding *ferritin*). The diploid potato clones with high and low levels of ACD from an ACD segregating family were used. Relative quantitative RT-PCR was used to study the differential gene expression. The differential gene expression analysis showed significant differences in some of the clones tested. The chlorogenic acid, citric acid and iron concentration were measured for the selected potato clones. The chlorogenic acid and citric acid were measured using HPLC and iron concentration was measured using atomic absorption spectroscopy. The detailed statistical data will be presented.

Effectiveness Of Using Native Polyacrylamide-Gel-Electrophoresis In Detecting Potato DNA Polymorphism

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Detection of DNA polymorphism is the basis for genotyping. Polyacrylamide gel electrophoresis (PAGE) is a commonly used approach in DNA polymorphism detection. The DNA fragment separation on denatured PAGE gels is based on the length difference between the fragments. Although the native PAGE is also mainly based on length difference of DNA fragments, it may also detect other types of differences such as conformational differences between DNA fragments. It has been reported that some DNA fragments can give apparent molecular sizes up to 10% larger in native PAGE than the fragments actual sizes. However, little is known about the extent of such kind non-length-based DNA polymorphism detected by native PAGE gels in potato DNA analysis.

We used denature PAGE gels, native PAGE gels, and agarose gels to compare the DNA fingerprints patterns of potato cultivars after amplification with polymerase chain reaction (PCR). Gels were stained with ethidium bromide and photographed using a gel documentation system. The number and molecular size of DNA bands of the same DNA samples were scored from different gels. The DNA primers in PCR reaction are mainly from the simple sequence repeat regions (SSR) of the potato genome.

Majority of the DNA primers showed additional DNA bands on native PAGE gels compared to the bands observed on denatured PAGE gels. The evaluated molecular sizes of most DNA fragments are quite similar between denatured PAGE gels and agarose gels. However, the apparent size of a fragment can be several-fold larger on native PAGE gels than its actual size. The results suggest that native PAGE can effectively detect both length polymorphism and non-length-based sequence polymorphism. The non-length-based polymorphism was found quite often in PCR-products amplified from SSR regions of the potato genome in current study. This information may contribute to the experimental design of gel electrophoresis for an effective detection of DNA polymorphism in DNA fingerprinting, DNA-based diagnostics, and genetic mapping.

Insecticide Resistance in Potato Pests: A Molecular Approach

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Quick, reliable and effective methods to distinguish insecticide resistant pests from susceptible ones and to determine dynamics of the resistance play an important role in pest control and in prevention of selection for high resistance levels. In order to monitor insecticide resistance in Colorado potato beetle (CPB) and potato aphid, we developed a Real-time PCR based method to distinguish Azinphosmethyl-resistant from susceptible Colorado potato beetles. Resistant or susceptible homozygotes were detected only by resistant (Res) or susceptible probe (Sus), respectively while heterozygotes were detected by both. The method can be potentially used for monitoring dynamics of Azinphosmethyl resistance in field CPB population. Insecticide resistance in insect is caused by mutation in specific genes such as nicotinic acetylcholine receptor genes and P450 cytochromes genes. Cloning of nicotinic acetylcholine receptor genes that is responsible for resistance to neonicotinoids and cloning of P450 genes that may relate to resistances of CPB and potato aphid, are also conducted. Six P450 genes have been discovered in CPB and one in potato aphid using degenerate PCR. Further screening for P450 genes and characterization of the discovered genes are still underway. Through comparison of the genes between resistant and susceptible populations and elucidation of the associated genes directly and indirectly with resistance we hope to find gene markers for insecticide resistances. Thus, we can use the markers to monitor dynamics of the insecticide resistance in insect population.

Identifying Potato Fields at Risk of Colonization by the Adult Colorado Potato Beetle

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An Integrated pest management program for Colorado potato beetles becomes fully effective only when both preventative and curative insect control methods are brought together. Frequently, IPM is limited to the regular monitoring of the CPB population to determine if the insect has reached an economically destructive level of abundance in potato fields. Then, a decision is made on the selection of a synthetic or biological insecticide and on the timing of a spray application to cure the problem. Less frequently, the risk of spring colonization by the adult Colorado potato beetle is assessed in fields considered for potato production. Then, fields at high risk of colonization can be protected using a soil systemic insecticide or by postponing their use for potato.

Current insecticide usage against the CPB on potato is reactive. Farm managers must be able to anticipate or forecast the relative abundance of the CPB in different fields if they are to plan their control practices so as to minimize insecticide requirements and to maximize the use of preventative alternative control methods including crop rotations. The tools to forecast colonization risks in potato fields are missing or not adapted to Atlantic Canada. The objective of the research introduced in this presentation is to use recent data and information on crop colonization by CPB to develop a model of crop invasion at a local and regional scale.

The presence of overwintering adult CPB in overwintering sites and along the edges of the emerging potato crop were monitored in 10 commercial fields located in the Jacksonville and Avondale areas of New Brunswick in 2004. The absence of overwintered beetles in grassy, bare soil or roadway habitats but their presence in most of the treed habitats support the concept of "privileged" overwintering sites and suggest that the location of dispersal points for field colonizing beetles in the spring could be mapped from a survey of the land-habitats surrounding these new potato fields. Even at the very low CPB density experienced in 2004, colonization of fields was detectable and occurred over an extended period of time. The location of the 2004 potato fields adjacent to or within 0.5 km of the 2003 potato fields seems to have created a landscape favorable to the persistence of a regional population of CPB.

The data have been used to develop a simple predictive model of field locations most at risk of colonization by adult CPB. Further experiments are necessary to parameterize the model

and test the validity of the assumptions.

Assessment of Efficacy of In-Furrow and Foliar Insecticides for Controlling Colorado Potato Beetle and European Corn Borer

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In recent years the European Corn Borer (ECB), *Ostrinia nubilalis*, has been elevated from a secondary potato pest to its current status as a primary pest in potatoes in the Maritime region. The common cultural practice of using in furrow imidacloprid, while currently providing effective control of Colorado Potato Beetle (CPB) and flea beetle, has not proven to be effective in controlling ECB larvae.

The project was conducted in 2004 in randomized block research plot islands at Cavendish Farms, New Annan, PE. Two varieties were used in the trials, Russet Burbank and Shepody planted at 18" and 12" respectively. Four replications were planted for each insecticide treatment. Plot islands consisted of two twenty-foot rows of each variety. The replicated trial was designed to assess the efficacy of two foliar insecticides, Success and Novaluron, for CPB control.

Treatments were as follows:

Check (no insecticide)

Admire In-Furrow (Standard Practice)

Foliar Success June 29 & Foliar Admire July 26

Foliar Success June 29 & Foliar Success July 26

Foliar Novaluron July 7 & Foliar Admire July 26

Foliar Novaluron July 7 & Foliar Novaluron July 26

Foliar insecticide products were applied at the following rates; Success 60ml/acre, Novaluron 350ml/acre, Admire 80ml/acre; timed to CPB larvae observations. Admire IF gave excellent suppression of over-wintered adult CPB (spring feeding activity), and completely controlled larvae. In the foliar insecticide treatments, a limited amount of feeding activity was observed from adult over-wintered CPB and slight feeding activity by larvae. However, all four foliar treatments gave adequate control of CPB larvae. Very extensive defoliation occurred in the check plots from the CPB larvae.

Given that the different foliar treatments gave effective control of CPB, Admire IF was considered the check for ECB observations. Application of foliar insecticides was timed for control of CPB larvae and not the hatching of ECB. ECB egg laying was not observed in the plots until after the first application of insecticide. Novaluron and Success were not applied

on the same dates, thus no direct comparisons of efficacy may be made between the two products. However, the four foliar treatments may be compared to Admire IF.

Plots were assessed by 3 methods: visual survey of stalk breakage; ratings of incidence, severity, and intensity of infestation; and by grading to Cavendish Farms processing contract.

On September 7th, the plots were visually assessed for stalk breakage on a scale of 1-10, zero to complete stem breakage respectively. The visual assessment showed that the Admire IF plots sustained much greater stalk breakage due to ECB damage than the plots treated with foliar insecticides regardless of variety.

On October 7th and 8th, ten plants of each variety from the inside row of each plot island, for a total of 40 plants per treatment were examined for: number of stems infected, number of holes per plant, and number of larvae. Expressed as incidence (% of stems infected), severity (#holes/infected stem) and infestation (#larvae/infected stem) the results were as follows. Average incidence for the treatments showed that there was high natural pressure from the ECB, with a range of 58% (Novaluron and Novaluron) to 94% (Admire IF). Shepody had a higher incidence than Russet Burbank. Results for severity and infestation in both varieties followed the same pattern with dramatic reductions in the infestation levels for the foliar insecticide treatments.

On October 8, these same rows were harvested to be graded to the Cavendish Farms processing contract. Grading included assessments of tuber size, fry colour, specific gravity and internal defects. Both of the Novaluron treatments had the highest contract payables for both varieties; the Admire IF had the lowest payable. This is attributable to the Novaluron plots attaining the highest biological yields, least smalls, and higher specific gravities. The higher ratings and grades for the Novaluron treatments could be attributed to the greater integrity of the photosynthetic and translocation systems in the plants with the least stem damage.

What has precipitated the elevation of the European Corn Borer to a primary potato pest in recent years? Possibly there is a correlation between the rise in ECB and the increased use of Admire IF for CPB and flea beetle control with a corresponding decrease in the use of foliar insecticides (J. Coffin, Fruit and Veg. Mag. July/Aug 2004). Our study showed that application of foliar insecticides aimed at CPB larvae significantly decreased the number of ECB. As the ECB larvae do not spend prolonged periods of time outside of the stem, and egg laying and hatching occur over an extended time frame, it is desirable for the insecticides to have a lengthy residual time to have maximum efficacy.

The Effect of Timing and Frequency of Insecticide Applications on ECB Infestation and its Impact on Russet Burbank Potatoes

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First reported in North America in 1917 in Massachusetts, the European Corn Borer, *Ostrinia nubilalis*, has rapidly expanded its geographical range ever since. Maine reported high populations in 1983 in the Houlton area but the population steadily declined until 1992 then began to rise again in 1995 (Dwyer 1999). On Prince Edward Island, damage on potatoes by this pest was not noted until 1987 (Stewart 1991). In New Brunswick, European Corn Borer populations became a concern in 1999 in Florenceville and in 2001 in Grand Falls. The population has been on the rise and has reached what is believed to be an economic threshold in some fields. Very little is known about the impact of the European Corn Borer on potatoes grown in northwestern New Brunswick. This study was initiated to determine the most appropriate spray schedule and quantify the effects of this pest on the late season processing cultivar Russet Burbank.

The trial was a completely randomized design with 4 replicates. The crop was planted May 24 following a one-year grain rotation. The area where the field was located has been a 'Hot Spot' for ECB activity since 2001. Plots were scouted for ECB eggs starting in mid-July by examining 20 plants randomly selected in the control plot. Development of egg masses was monitored and first insecticide application was made at first sign of egg hatch.

Treatments were as follows:

- 1) Control, no insecticide (C)
- 2) One application of Success at first sign of egg hatch - July 22 (1E)
- 3) One application of Success one week after 1E - July 28 (1M)
- 4) Two applications of Success – July 22 and 28 (2E)
- 5) Two applications of Success – July 28 and August 4 (2M)
- 6) Three applications of Success – July 22, 28 and August 4 (3)

Incidence and severity of crop damage was rated on September 13 to 15 by examining one stem each of 10 plants randomly selected in each of the four replicates and six treatments for a total of 40 stems per treatment and 240 stems for the experiment. Number of stems infected and total number of holes were counted. On October 14, treatments were mechanically harvested. The total number of plants, stems and tubers were counted. The crop was graded according to contract.

ECB infestation incidence, % stems infected, was 100, 100, 100, 100, 98 and 93 percent while infestation severity, number of holes per stem infected, was 27.3, 10.1, 12.3, 9.4, 6.7 and 5.0 for the C, 1E, 1M, 2E, 2M and 3 treatments respectively. Incidence of infestation was not significantly reduced by insecticide applications while severity of infestation was significantly reduced by all insecticide treatments. Plots receiving an insecticide application on August 4, treatments 2M and 3, had significantly less holes per stem than treatments 1E, 1M and 2E. Total and marketable yield was significantly increased by the first but not the second or third insecticide applications, but there was little effect of timing and frequency of insecticide application. Controlling ECB improved specific gravity but differences were only significantly higher in treatments receiving two and three insecticide applications: 2E, 2M and 3.

Under the conditions experienced in 2004, Russet Burbank's yield was not significantly affected when 93 to 100% of the stems had 5 to 12 ECB holes. Reducing infestation severity to between 5 and 9 holes per stem significantly improved specific gravity over the control where stems averaged 27 holes. Another variety or even the same variety grown under a different rotation, in another growing season could respond differently.

The challenge in successfully implementing a European Corn Borer on-farm Integrated Pest Management program resides in the understanding that threshold levels will vary depending on many factors such as variety, fertility, moisture, timing of insect infestation, interaction with other pests and diseases, season and possibly others. At best, thresholds represent "educated estimates" and should be regarded as being in the "ball park".

European Corn Borer in Potatoes and a Possible Biological Control

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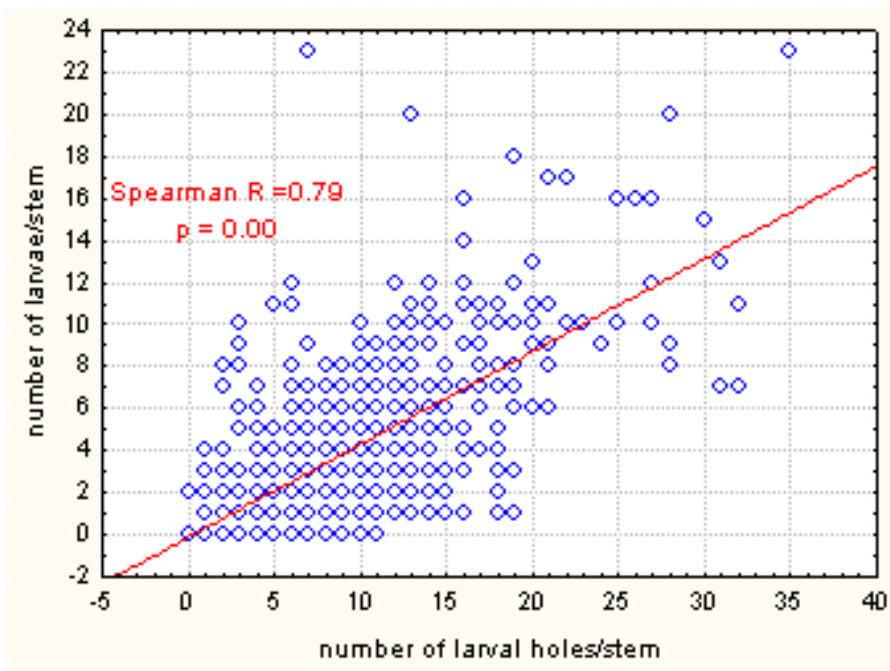
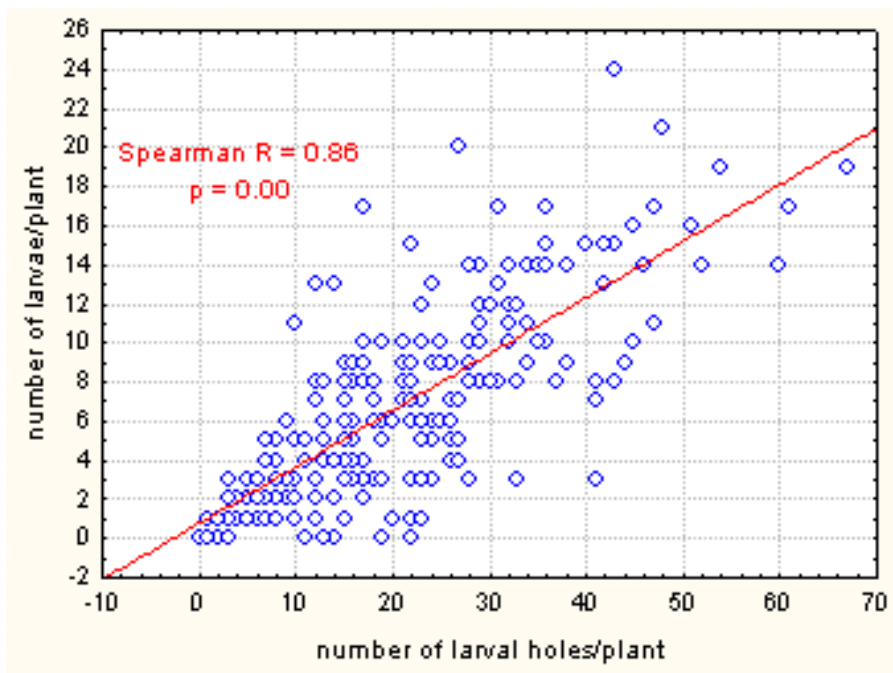
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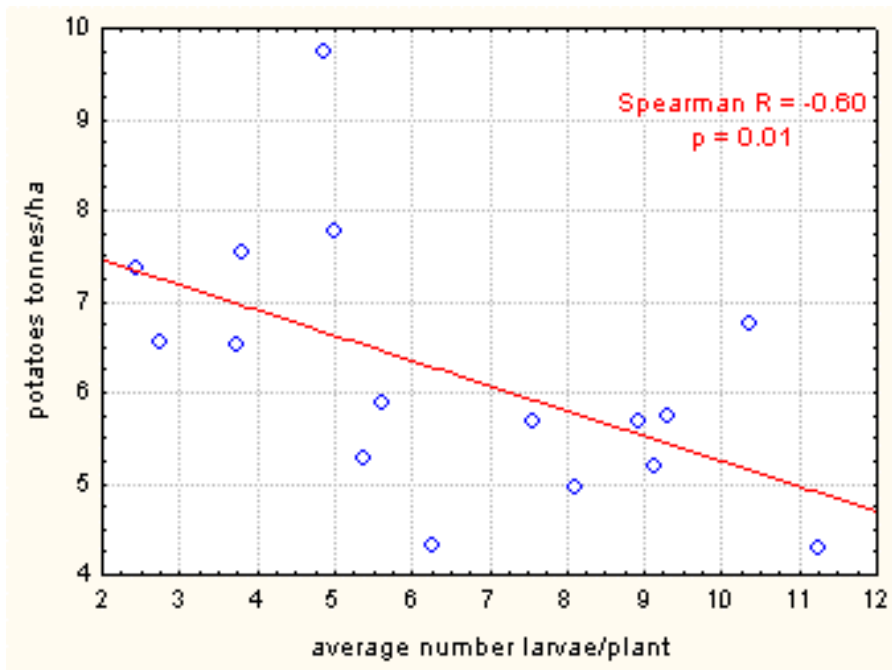
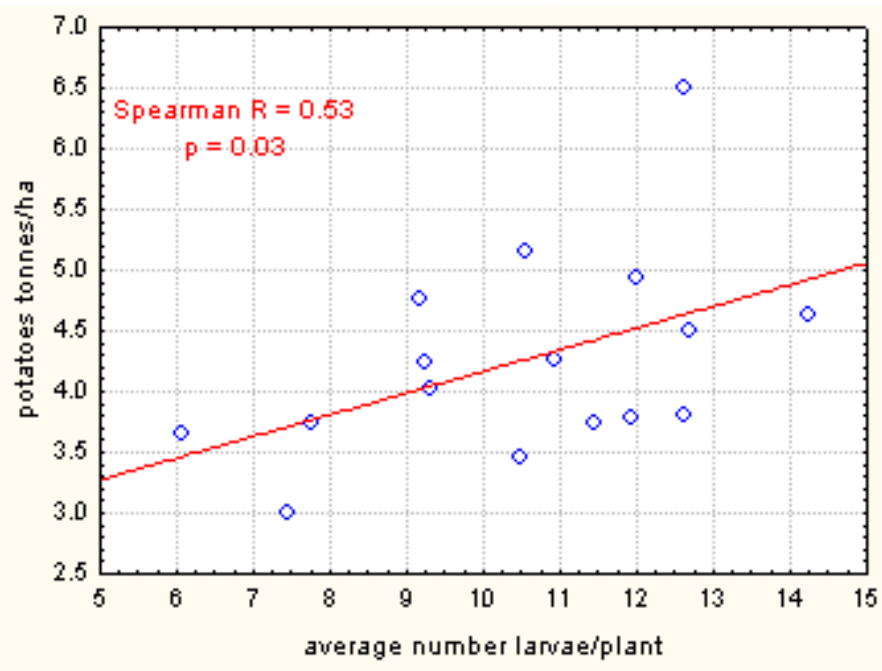
European corn borer (ECB) is an emerging potato pest in Atlantic Canada and Maine which, because of its ability to bore into the potato stems, is difficult to quantify during infestations and difficult to control because many insecticides are only effective during the few days between egg hatch and larval entrance into the potato stem. As well, while the ECB damage of potato stems is well documented the response of the potato plants to this damage with regards to tuber yield is mixed.

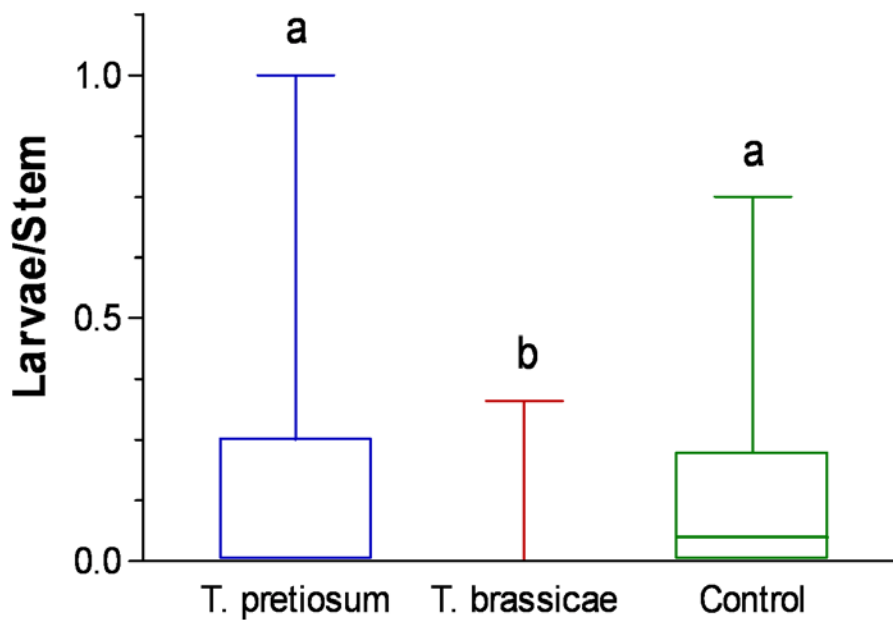
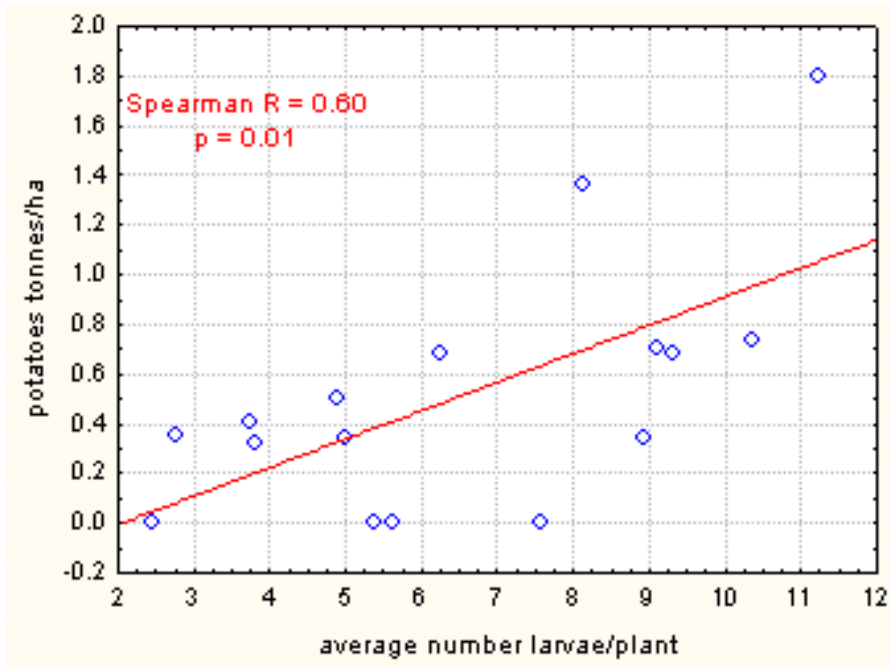
At the end of the 2004 growing season a total of 3085 stems were collected from potato plants in 55 plots set up in potato fields as part of a variety of research projects. Each of these stems was sliced open and the number of ECB larvae, larval holes, and larval tunnels was recorded for each individual stem and plant. The results of this work, showed a significant correlation between the number of larval holes per plant and the number of ECB larvae per plant (Figure 1). There was also a significant correlation between the number of larval holes per stem and the number of ECB larvae per stem (Figure 2). This data shows that the number of larval holes will give an excellent indication of the number of larvae in either the potato plant or the potato stem.

One of the research projects mentioned above included 16 test plots of Shepody potatoes and 16 test plots of Russet Burbank potatoes. Both sets of test plots showed essentially no correlation between the number of larvae per plant and total potato yield. The Shepody plots, however, showed a significant positive correlation between the number of ECB larvae per plant and the total weight of Canada #1 small potatoes (Figure 3). The Russet Burbank plots showed a significant negative correlation between the number of ECB larvae per plant and the total weight of Canada #1 small potatoes (Figure 4) and a significant positive correlation between the number of ECB larvae per plant and the total weight of Canada #1 large potatoes (Figure 5). The difference in the response to ECB damage was probably a result of the difference in the growth type of these two varieties.

The parasitic wasp, *Trichogramma*, has been used to successfully control ECB in corn in many areas of the world (Smith, 1994). The *Trichogramma* females lay their eggs in insect host eggs and the wasp larvae consume the contents of the host egg as they complete their life stages. The use of *Trichogramma* to control ECB in potatoes was investigated using two species, *T. brassicae* and *T. pretiosum*. Potato plants treated with *T. brassicae* showed a significant reduction in ECB larvae (Figure 6) as well as in larval holes and larval tunnels. These results indicate the need for further investigation.







European Corn Borer Research in PEI – 2004

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Randomized plots were established in two commercial Russet Burbank potato fields to determine the effects of one and two spray applications of spinosad (Success) vs. an untreated control on European corn borer populations. The number of egg masses per plant in each plot was recorded in mid July and growers were asked to spray when flagged egg masses reached the 'black head' stage. A second spray application was made one week later in the plots receiving two applications. Within one week of topkill, damage (based on number of holes per stalk) and larval counts were recorded for each plot. Plots were harvested and marketable yields and grades were determined according to processing requirements.

Both the one and two spray spinosad treatments significantly reduced the number of larvae per stalk and damage when compared with the untreated check. There was no significant difference in yield (total or marketable) even with larval infestation rates as high as one larvae per stalk and up to four holes per stalk in the untreated plots. There was also no significant difference in crop value for processing when looking at quality aspects like specific gravity and percentage of tubers over ten ounce.

Research will continue this growing season to determine if these trends are consistent from year to year. It is suspected that environmental conditions such as available moisture may affect the severity of damage seen in the field.

Real-Time PCR Quantification of Denitrification Genes in Bacterial Strains and Potato Field Soils

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Denitrification is a biological process resulting in the reduction of nitrate and nitrite to yield nitrous oxide and molecular nitrogen (see Figure 1). Denitrification has environmental significance both as an important sink for NO_3^- and NO_2^- , potential water contaminants, and as a source for N_2O , an important greenhouse gas and ozone depleting substance. From an agronomic perspective, denitrification results in the loss of plant available nitrogen from the soil. Whilst a number of factors are known to influence denitrification rates in the field (ie landscape characteristics and land management, organic carbon content, soil bulk density, pore size distribution), it has been difficult in the past to study the microbial population that is responsible for denitrification.

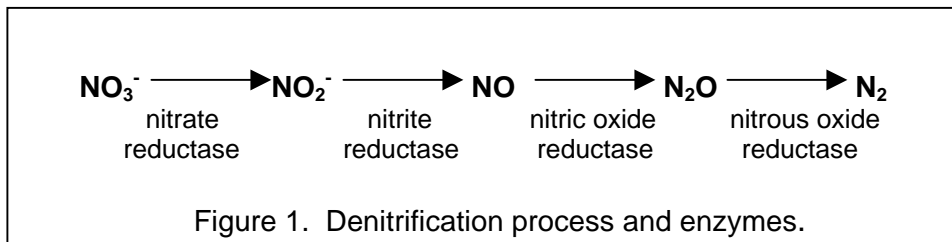


Figure 1. Denitrification process and enzymes.

Historically, denitrifying bacteria have been isolated from a range of environments such as agricultural soils, marine sediments and wastewater treatment plants. The genes encoding enzymes involved in the denitrification process have been cloned and sequenced from a number of these strains (Phillipot, 2002). The denitrification process utilises 4 major enzymes – nitrate reductase nitrite reductase, nitric oxide reductase and nitrous oxide reductase. These enzymes are coded for by 7 major genes – *napA* and *narG* (nitrate reductase), *nirS* and *nirK* (nitrite reductase), *cnorB* and *qnorB* (nitric oxide reductase) and *nosZ* (nitrous oxide reductase). Studies on denitrification tend to focus on the steps from nitrite reductase onwards, as nitrate reduction can occur without the production of gaseous emissions. It is known that there are many more bacteria in environmental samples than can be cultured using current techniques, so molecular biology methods have been utilised to study the diversity of gene sequences of denitrification genes (Braker *et al.*, 1998; Braker and Tiedje, 2003; Scala and Kerkhof, 1998). Using this sequence information, our aim is to generate primers for PCR that will enable us to amplify DNA from the denitrifying population

in potato field soils.

Real-Time PCR is one recent technique that enables quantification of target copies of a DNA sequence. Real-Time PCR quantifies the amount of product obtained during each cycle of a PCR reaction, by use of a fluorescent probe or intercalating dye. Copy number of the target DNA is determined by comparison with a standard curve of known concentrations. Real-Time PCR is specific, accurate and can quantify targets over a large dynamic range and is therefore considered the method of choice for quantitation of target sequences in environmental samples (Gruntzig *et al.*, 2001).

Studies have commenced to determine appropriate molecular tools for quantification of denitrification genes using Real-Time PCR. Initial studies have focussed on utilising available primers/probe for quantification of nitrite reductase gene (*nirS*) of *Pseudomonas stutzeri* (Gruntzig *et al.*, 2001), a well-characterised denitrifier that is commonly isolated from soil environments. Preliminary results have confirmed the specificity of the detection system for *P. stutzeri* (by testing against several denitrifier species) and the ability to quantify target copies over a large dynamic range (see Figure 2). Studies are ongoing to quantify the population of this target in potato field soil. Due to the very specific nature of the *P. stutzeri* primer/probe set, primers have also been designed to try to encompass a larger range of nitrite reductase (*nirS*) genes/clones from the database. One set of primers is able to amplify *nirS* gene fragments from *P. stutzeri*, *Azoarcus tolulyticus* and *Alcaligenes eutrophus*.

Initial testing has also commenced to target the range of other genes involved in the denitrification process (ie nitrite reductase gene (*nirK*), nitric oxide reductase gene (*norB*) and nitrous oxide reductase gene (*nosZ*). Published primers are available for *nirK* (Henry *et al.*, 2004), but initial testing indicates that specificity of these primers may be a problem under our PCR reaction conditions.

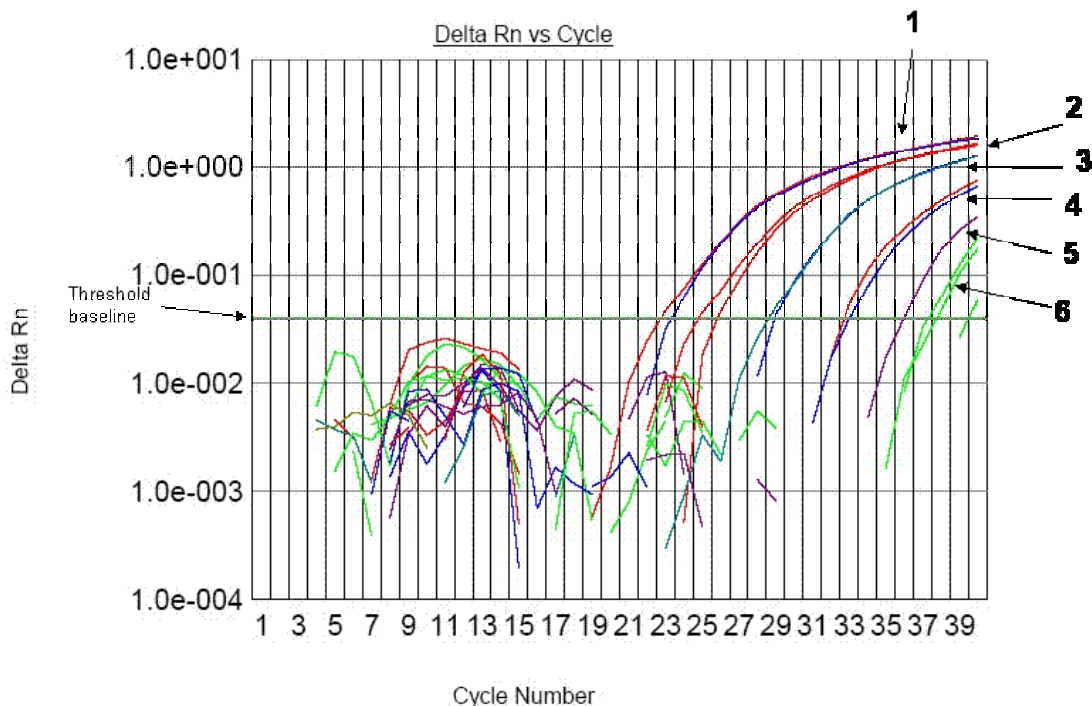


Figure 2. Real-Time PCR amplification curves using varying template concentrations of *P. stutzeri* ATCC 14405 genomic DNA template. 1 = 20ng, 2 = 5ng, 3 = 0.5ng, 4 = 50pg, 5 = 5pg, 6 = 0.5pg. Horizontal line indicates threshold baseline used for calculating C_T (threshold cycle) for quantitation of target copy number.

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Effect of Soil Type and Nutrient Management on Potato Tuber After-cooking Darkening

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Potato after-cooking darkening (ACD) is one of the most widespread, undesirable, characteristics of potato tubers. It has been reported from every potato growing area worldwide, and is one of the important quality indicators in table and processing potato varieties. ACD has been known for its strong genetic control and the strong impact of certain environmental factors. No single factor or combination of factors is uniquely responsible for the ACD. Genetic control governs darkening more than any other factors, but the tendency of potato tuber to darken after cooking also depends on the plant growth environment, including soil type and its fertility, moisture content, temperature, tuber maturity at harvest, and the length and conditions of storage. A combination of these factors determines the degree of the pigmentation, which ranges from light gray to almost black.

Soil fertility determines the darkening level of the potato tubers. Early research showed evidence that high organic matter and high nitrogen increase ACD in tubers, and high amounts of potassium and nitrogen fed to plants reduce ACD. Iron content in tubers, on the other hand, depends on soil type and growth conditions. This study used the processing variety Russet Burbank as a model to investigate the effect of management practices and climatic conditions on ACD, focusing on soil properties (texture and organic matter), fertilizers (nitrogen, phosphorus, and potassium) and the rates of fertilizers. The effect of these factors on ACD during tuber storage was also investigated. The statistical analyses have shown significant impact of soil type, soil and nitrogen interaction and interaction of soil with other fertilizers to ACD. Detailed statistical analyses will be presented during the meeting.

Weed Control in Potatoes using Physical, Thermal, and Organic Products

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More effective weed control methods are needed in Canada for potatoes (*Solanum tuberosum* L.), that are grown with reduced chemical inputs. We evaluated several non-chemical techniques including physical methods (mechanical, thermal, banding), use of acceptable organic alternatives (corn gluten and acetic acid), and weed emergence programs, used alone and in combination, and compared to a herbicide treatment for effectiveness in managing weeds.

Field experiments were conducted over several years at Charlottetown, Prince Edward Island on a fine sandy loam soil with a pH and organic matter from 5.8 to 6.0 and 2.6% to 3.0%. Plots were set up in plots 4 rows wide in randomized complete block design with four replications. Fertilizer was applied in the physical methods and acetic acid experiments in a band 5 cm below and to the side of the row as 15-15-15 (N-P-K) at 950 kg/ha at planting. Russet Burbank seed pieces were machine planted between May 18 and 30 with 45 cm in row spacing and 0.9 m between rows with rows 6.0 to 8 m long. The gluten experiments were fertilized with 30 t/ha of compost and were planted between June 1 and 5 to Superior cultivar with 25 cm in row spacing and same design as with Russet Burbank. Treatment effects on grass and broadleaved weeds were assessed using weed fresh weight. Yields were obtained at maturity by mechanically harvesting two plot rows and grading into marketable and total tubers. Data collected was subjected to the analysis using ANOVA and LSD was used to separate the mean differences.

Physical methods and gluten: The physical methods examined in 2002 and 2004 were basket weeder, finger weeder, and power tiller, and banded over the row treatment of herbicide, flamer, or gluten in combinations with the physical methods. Aggressive mechanical methods achieved good weed control between potato rows and when coupled with effective in-row systems effective weed control with high potato yield was obtained. Averaged across tillage treatments, the power tiller gave weed control comparable to use of a herbicide between the rows, the basket weeder gave less but acceptable weed control, and the finger weeder was less effective. Thermal and herbicide gave excellent and comparable weed control when applied in 25 cm bands over the row; however, the gluten was not effective but did give positive effects on potato tuber

diseases. Marketable yields were lower where gluten was used due to poor weed control.

Acetic acid: Acetic acid was evaluated in 2004 as (a) the commercial formulation of Ecoclear applied at 800 L/ha pre-emergence to the potatoes, at ground crack and post-emergence and as (b) glacial acetic acid at three concentrations of 10, 20 and 30 % solutions applied pre-emergence only or twice at pre-emergence and at ground crack. Ecoclear gave excellent control of wild radish, corn spurry and lamb'squarters by 6 weeks after application. Ecoclear caused damage to the potatoes when used post-emergence but marketable and total yield was not affected. Glacial acetic acid at 30% gave best weed control with lower concentrations being progressively less effective. The 2x application was most effective. All treatments of acetic reduced potato marketable yield slightly compared to a herbicide treatment.

Weed Emergence Timing: The Weedcast software program was used in experiments in 2001 to 2003 as a method to predict the most appropriate time to cultivate potatoes to remove lamb's-quarters. Weeds were removed by cultivation each the program predicted lamb's-quarters emergence to be at 15, 30 or 60% emergence after potato planting. As a result, plots were cultivated 3x, 2x and 1x at the predicted emergence times of 15, 30 and 60% emergence, respectively. These plots were paired to plots treated with metribuzin + metolachlor and cultivated at the same time the Weedcast predictions. Addition of cultivation after herbicide application had very little effect on potato yield. One or two cultivations used alone, did not provide sufficient weed control and as a result yields were reduced to levels below that obtained with herbicides. Three cultivations gave good weed control and yields were comparable to the herbicide + cultivation treatments in one of three years. Timing cultivation at the 15% predicted emergence of lamb's-quarters using Weedcast appeared to be a useful tool for weed management. Weed management and resulting potato yield were very much affected by yearly weather and weed density when cultivation was sole method of weed control used but had little effect of results when herbicides were used to control the weeds.

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How Can Field Terracing Impact on Nutrient Management in Potato Production

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Diversion terraces combined with grassed waterways are efficient soil and water conservation practices in a potato field. Subsequent changes from conventional up-and-down tillage practice to cross-slope tillage method parallel to the diversion ditch reduce both the water runoff velocity and soil and nutrient losses. This study tried to monitor the effects of such changes on the distribution of soil nutrients and potato yields on the terrace and the landscape. A terraced field (5 terraces) covering 14.4 ha and subdivided into two sections separated by a grassed waterway was selected. The grassed waterway drained the terraces and ditches to a wood lot at the end of the field. The field was cropped to grains in 2003 and potatoes in 2004. After the grain harvest, soil samples were collected based on a 50m by 60m grid (the terrace width). A composite sample was taken from random samples collected inside each grid unit. The grassed waterway and the wood lot were also soil-sampled. All the grid corners were georeferenced. Further details on the distribution of soil nutrients on the terrace were collected every 6 m in the spring of 2004 along a baseline set across the terraces. Potato yields were monitored in the fall every four rows along the same baseline.

Ordinary kriging using the grid data did not show any landscape position effects on the distribution of soil nutrients across the terraces. The field edge effects were however detected for soil phosphorus. The lowest soil P levels were measured from a 50-m field section wide away from each side of the grassed waterway and at the edge section towards the wood lot. The sampling grid unit was probably too large to allow for any detection of the soil variability as a result of landscape position and terracing. Soil nutrient levels, particularly soil phosphorus, were very high (> 78 ppm Mehlich-3 P) including in the waterway soil. Soil P levels varied from 51 to 87 ppm P in the wood lot. Such P enrichments resulted from the export of nutrients from the terraces. Across the terraces, the results confirmed the edge effects at the bottom section of the watershed. Moreover, they detected the edge effects along the diversion ditches. Overall, sinusoidal-like functions associated with linear and quadratic trends appeared to characterize the increase of soil nutrients and potato yields from upside to downside of each terrace. More specifically, soil P levels and P saturation percentages alternatively reached two minimum and two maximum values from the upside to downside of the terrace. Potato total tuber yields and quality followed similar patterns. Such variations might have resulted from a combination of soil compaction (due to tire tracks) and

water and nutrient transfer factors (water and tillage erosion) on the same terrace.

In conclusion, the study results tended to confirm important differences in soil quality and potato yields exist across the same terrace while no or small difference exists between terraces. As a result, nutrient management on the terrace can be adjusted, accordingly. Further investigations are however needed before a final conclusion can be made.

Alternative Biological Amendments: Effects on Soil Biology and Soilborne Diseases

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Soilborne diseases of potato can be persistent, difficult-to-control problems in potato production, and typically result in substantial losses in tuber yield and quality. Although fungicide sprays and seed treatments can provide some control for certain diseases, these treatments are not always practical or effective, and particular soilborne diseases remain recurrent problems. In addition, reducing pesticide inputs and achieving more integrated, sustainable disease management approaches are desirable. One approach with potential to control multiple soilborne diseases that has received increasing attention and interest in recent years is the use of biological amendments. Biological amendments can include a wide variety of treatments, including governmentally-registered biocontrol agents, diversified mixtures of beneficial microorganisms (microbial inoculants), mycorrhizae, composts and compost teas, and growth-enhancing extracts, enzymes, organic acids and natural compounds (biostimulants). The goal of all biological amendments is to manipulate or alter the soil microbial characteristics through the addition or stimulation of beneficial microorganisms that will result in suppression of diseases by increasing microbial activity, diversity, and antagonism towards pathogens. Addition of biological amendments may affect microbial communities by displacing, suppressing, or inhibiting particular microorganisms, or by stimulating others, or through changing the microbial environment in ways that affect other organisms. These added microorganisms may also be negatively affected by the already established soil microflora, and they must be capable of establishing, surviving, and persisting at population levels necessary for adequate disease control.

Over the last few years, our lab has conducted field and greenhouse evaluations of numerous biological amendments (Table 1). Research results indicate that most biological amendments are successful at introducing the beneficial microorganisms into the soil environments, at least initially. Soil fatty acid (FAME) profiles measured 2 weeks after application of biological amendments show shifts in microbial community characteristics of varying degrees and types depending on the specific amendment and application rate. However, these effects may be short-lived, and, in some cases, microbial effects could not be detected by the end of the field season.

The commercial biocontrol agents *Bacillus subtilis* (*Bsub*), *Burkholderia cepacia* (*Bcep*), *Trichoderma virens* (*Tvir*), and *Trichoderma harzianum* (*Tharz*), all significantly reduced stem canker and all but *Tharz* reduced black scurf, caused by *Rhizoctonia solani*, and *Bsub* and *Tvir* also increased total and marketable tuber yield, over multiple field seasons. Some treatments, including vitazyme (vita), Effective microorganisms (EM-1), and foliar-applied compost tea (C.T.) produced no significant effects on soilborne diseases, tuber quality, or tuber yields in our trials. In other cases, significant effects and trends, indicating reduced disease and increased yields, were observed for mycorrhizae (AMF) and soil-applied compost tea (C.T.) treatments, but these effects were not consistent. When used in conjunction with different 2-yr rotations, some treatments (such as C.T. and Mix+) showed significant disease control in some rotations, but not in others. Alternative biological amendments that effectively manipulate soil microbial communities, resulting in reduced soilborne disease and increased tuber yields, could play an important role in potato disease management strategies, but more research is needed to determine the most effective treatments, the degree, extent, and duration of efficacy, and economical means of implementation.

Table 1. List, description, and trade names of biological amendments evaluated

Abbrev.	Active agent(s), description	Product name
Biocontrol agents		
<i>Bsub</i>	<i>Bacillus subtilis</i> strain GB03	Kodiak
<i>Bcep</i>	<i>Burkholderia cepacia</i> type Wisconsin Isolate J82	Deny
<i>Tvir</i>	<i>Trichoderma virens</i> strain GI-21	SoilGard
<i>Tharz</i>	<i>Trichoderma harzianum</i> strain T-22	RootShield
Microbial inoculants		
EM-1	Activated mix of yeasts, lactic acid bacteria, and phototrophic bacteria	Effective Microorganisms
EM-B	Wheat bran anaerobically fermented with EM-1	EM Bokashi
C.T.	Aerobically-brewed compost tea (24 hr) from vermicompost	Earth Tea Brewer (22-gal)
VC	Vermicompost (worm castings) - source of compost tea	Vermicompost
AMF	Mix of arbuscular mycorrhizae, several <i>Glomus</i> and <i>Gigaspora</i> spp.	Endomycorrhizal Inoculant
Mix+	<i>Bacillus</i> , <i>Pseudomonas</i> , <i>Streptomyces</i> , and <i>Trichoderma</i> spp., + nutrients	Compete Plus
Biostimulants		
Vita	Vitamins, enzymes, organic acids, growth regulators, and plant extracts	Vitazyme
Chemical seed treatment		
TMC	Topsin/mancozeb/cymoxanil (TMC) as a seed treatment control	Evolve

Late Blight and Pink Rot of Potato: Disease Complex Situations

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Summary

Potato late blight, caused by *Phytophthora infestans* (Mont.) DeBary and pink rot, caused by *Phytophthora erythroseptica* Pethyb. are important oomycete diseases in potato-growing regions world-wide. Potato foliage and tubers can be infected by *P. infestans* while roots, stolons and tubers can be infected by *P. erythroseptica*. While the former results in a dry rot that can be slow-growing resulting in intact but diseased tubers at the end of the storage period, the latter involves a wet rot that can lead to total stored crop loss within days to weeks. When warm, wet fall conditions exist, pink rot is more prevalent but if late blight has also been present during the growing season and/or pre-harvest period, post-harvest tuber losses dramatically increase. To investigate the cause(s) of this situation and the potential for a synergistic interaction between these two pathogens, studies were conducted involving the introduction of spores of these two pathogens into soil as potato tubers were developing. Soil inoculations included both individual and combined pathogen zoospore suspensions. Below ground plant tissues were assessed for disease occurrence. More disease was found with combination inoculations than with individual pathogen inoculations. Further studies are underway to confirm the existence of additive or synergistic interactions between these two pathogens.

Introduction

Late blight of potato (*Solanum tuberosum* L.), caused by *Phytophthora infestans* (Mont.) DeBary, is the most devastating disease of potatoes in Canada and worldwide. Late blight is spread initially by infected tubers that are always visibly diseased and then by airborne spores (sporangia) of the pathogen that form on infected, healthy plant tissues (Platt 1994). The sporangia settle on and infect healthy potato leaves and stems or land on the soil and produce motile ('swimming') spores (zoosporangia) that move to and infect potato tubers. Sporangia have a short survival period (hours to days) and are produced asexually on disease lesions of infected leaves and tubers throughout the disease development period until the plant is dead normally in a few days or weeks. Infected tubers have a slow developing dry rot that provides over-winter survival for the pathogen. However, in the past decade, pathogen populations worldwide have been changing due to the

introduction of new, more aggressive strains and of the second mating type of the pathogen (Fry et al. 1993; Deahl et al. 1995, Goodwin et al. 1994, Goodwin et al 1996, Platt et al 2002). When both mating types (A1 and A2) combine (two disease lesions join), sexually produced oospores are formed and these are genetically distinct and have a long survival period (months to years). In addition, many of the new strains of the pathogen are resistant to mefenoxam, a fungicide that has been widely used during the growing season for control of late blight of potato foliage and tubers, and able to overcome resistant hosts (Daayf 2002, Gisi 1996, Peters et al 1998).

Pink rot of potato (*Solanum tuberosum* L.), caused by *Phytophthora erythroseptica* Pethyb., is an important disease in potato-growing regions world-wide (Vargas and Nielsen 1972). The disease is particularly severe in poorly-drained land, when warm, wet climatic conditions persist at time of harvest (Bonde 1938; Rowe and Nielsen 1986). Underground potato tissues such as roots, stolons, tubers, and basal stems can be infected by *P. erythroseptica*; root and stem infection can result in plant wilting and death while tuber infection leads to pink rot development prior to harvest and in storage. Tubers are usually infected via the growth of mycelium from diseased stolons, however, when adequate moisture is available and favourable temperatures occur, zoospores can infect tubers directly through buds or lenticels (Vargas and Nielsen 1972). Harvested, infected tubers develop a wet rot symptom that can quickly result in the complete disintegration of the potato tissue. Control of pink rot has been provided by mefenoxam (or its parent isomer metalaxyl) for many years. Potato producers in Canada utilizing this control option apply Ridomil products as foliar applications during tuber initiation and again 14 days later. However, many areas in North America have pathogen populations that have become resistant to mefenoxam (Peters et al. 2003).

In the past decade, late blight and pink rot incidences and damage have increased resulting in major post-harvest crop losses. However, these occurrences are not always related to the use of mefenoxam and presence of mefenoxam-resistant pathogen populations. In many instances, in fields where late blight was a significant problem, particularly during the latter half of the growing season, a rapidly developing wet rot condition occurred within a few weeks of storing the crop. Examination of these rotting tubers has revealed the presence of both *P. infestans* and *P. erythroseptica*. The relationship between late blight in fields of potatoes prior to harvest and significant post-harvest crop losses due to pink rot was investigated in controlled disease complex studies.

Materials and Methods

Isolates of *Phytophthora infestans* and *P. erythroseptica*

All isolates of the pathogens used for study inoculations were recovered from infected potato plants or tubers in Canada. Isolates used in the studies were characterized with standard biological and molecular techniques (Gerritson-Cornell 1985, Newhook et al.

1978) and confirmed by staff at Agriculture and Agri-Food Canada, Eastern Cereal and Oilseed Research Centre, Ottawa, ON.

Greenhouse studies

In replicated greenhouse studies with a randomized block design (5 reps), healthy seed tubers (cv Shepody) were planted in standard potting soil in 20 cm pots. Standard lighting, fertilizer and water regimens were followed. After about 8 weeks growth, a pathogen inoculum suspension (approx. 10,000 spores/ml) was applied (45 ml/pot) to the soil near the main stem. Inoculum suspensions were prepared by scraping spores from 10-14 day-old rye or V-8 juice agar culture dishes of *Phytophthora infestans* or *P. erythroseptica*, respectively. Both A1 and A2 mating types of *P. infestans* were used. Pathogen inoculation treatments included: non-inoculated controls; *P. infestans*; *P. erythroseptica*; and *P. infestans* plus *P. erythroseptica*.

For the remaining plant growth period (7-8 weeks), plant growth and foliar disease occurrence was assessed weekly. At harvest, the following plant tissues were assessed for disease symptom occurrence and severity: above and below ground stem parts; stolons; and roots. Tuber assessments included the occurrence of late blight, pink rot, scab, black scurf, silver scurf, total rot, aerial and healthy tubers. Data were analyzed by analysis of variance (Genstat 5, Release 3[1], The Numerical Algorithms Group Ltd., Oxford, UK) and when a significant treatment effect was found, the test of least significant difference (LSD, $P = 0.05$) was used to separate means.

Results

No foliar disease was observed and normal plant growth occurred during the study period. Similarly, no significant differences in disease occurrence on above-ground and below-ground stem parts were apparent. However, significant treatment differences in the incidence of rot on tubers, stolons and roots were observed (Tables 1-3). In all studies, non-pathogen control treatments had healthy tubers, stolons and roots.

In all studies, inoculation of soil with *P. infestans*, *P. erythroseptica*, and *P. infestans* plus *P. erythroseptica* significantly affected the occurrence of tuber symptoms of late blight, pink rot and total rot. However, pink rot symptoms were only observed in inoculated treatments that included *P. erythroseptica* while late blight symptoms occurred only with *P. infestans* inoculations. In 2001 and 2004, more pink rot occurred following inoculation of both pathogens than with just *P. erythroseptica* while in 2002 there was less disease found following inoculation with *P. infestans* plus *P. erythroseptica* than with *P. erythroseptica* alone.

Late blight tuber rot symptoms were present in plots inoculated with *P. infestans* or *P. infestans* plus *P. erythroseptica* in 2001 and 2002 but not in 2004. In 2001 and 2002, the A1 mating type of *P. infestans* plus *P. erythroseptica* had similar disease levels as inoculations

with only the A1 mating type of *P. infestans* while in 2004, significantly more disease occurred with the combination treatment. For inoculations of *P. infestans* - A2 mating type, or *P. infestans* - A2 mating type plus *P. erythroseptica*, no significant differences among the treatments occurred in 2002 and 2004 while in 2001, significantly less disease was observed with the combined pathogen inoculations than with *P. infestans* – A2 mating type alone.

Total tuber rot levels were the same as or greater than pink rot or late blight tuber rot levels in all studies. In addition, total tuber rot tended to be the highest with combined pathogen inoculations in studies except for the *P. infestans* – A1 mating type plus *P. erythroseptica* treatment in 2001.

For stolon rot, no significant differences among the pathogen inoculation treatments were found in 2004 but in 2001 and 2002, more disease occurred with *P. erythroseptica*, and *P. infestans* plus *P. erythroseptica*. In some cases, the combined pathogen treatment had more disease than with solo pathogen treatments; e.g., *P. infestans* versus *P. infestans* plus *P. erythroseptica* in 2002. For root rot in 2002 and 2004, significantly more rot was observed in *P. erythroseptica* and *P. infestans* plus *P. erythroseptica* inoculation treatments. In some cases, the combined pathogen treatment had more disease than with solo pathogen treatments; e.g., *P. infestans* versus *P. infestans* plus *P. erythroseptica*. In 2001, no significant differences among pathogen inoculation treatments were found. In all studies, some stolon and root samples exhibited rot symptoms following inoculation with either pathogen.

Discussion

Potato tuber late blight and pink rot are an increasingly major source of post-harvest crop losses in North America and are frequently found to occur together. Fields of potatoes with foliar late blight caused by *P. infestans*, particularly late in the growing season, often have significant late blight tuber rot levels in the harvested crop. As late blight tuber rot is most often a slow developing dry rot, it can be successfully managed during the storage period with proper removal of visibly diseased tubers and management of the conditions (air temperature, humidity and ventilation) of the stored tubers. When weather and harvest conditions favour the occurrence of pink rot caused by *P. erythroseptica*, significant post-harvest crop losses can also result due the rapidly developing wet rot caused by this disease. In recent years, in conjunction with changes in pathogen populations, major post-harvest crop losses have occurred and these have been associated with the presence of both late blight and pink rot symptoms and pathogens.

To improve the management and prevention of post-harvest crop losses, it is necessary to more fully understand the roles and impacts of both *P. infestans* and *P. erythroseptica*. In greenhouse studies, inoculation of soil with solo or combined spore suspensions of *P. infestans* and/or *P. erythroseptica*, demonstrated that disease symptoms of late blight, pink rot or both were present on tubers and that root and stolon rot symptoms

were observed. Furthermore, in most cases, the combined pathogen inoculation treatments caused more disease than individual pathogen treatments. This suggests that foliar late blight and the subsequent occurrence of late blight tuber rot may predispose tubers to the development of pink rot. Unlike, the slow developing, easily managed post-harvest dry rot manifested by late blight tuber rot, the presence of both late blight and pink rot in the harvested crop often results in a rapidly developing wet rot that is very difficult to manage successfully.

While, these studies have demonstrated an additive and/or synergistic relationship between *P. infestans* and *P. erythroseptica*, more studies are needed to confirm the results and investigate this situation under field conditions. In addition, investigations are needed to determine the cause of the root and/or stolon rots following inoculation with *P. infestans*. Finally, studies are required on methods to minimize the risk and impact of the combined pathogen situations prior to and after harvest to more successfully prevent and manage these damaging post-harvest crop diseases.

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Table 1. Tuber, stolon and root rot symptom incidences (mean numbers per rep) following solo and combination inoculations of *Phytophthora infestans* (Pi) and *P. erythroseptica* (Pe) in 2001.

Soil Drench Inoculation Treatment	Tuber Pink Rot	Tuber Late Blight	Total Tuber Rot	Total Stolon Rot	Total Root Rot
Control	0	0	0	0	0
Pi, A1 mating type	0	0.6	0.6	0.2	0.2
Pi, A2 mating type	0	1.4	1.4	0	0
Pe	0.2	0	0.4	0.6	0.4
Pi-A1 + Pe	0.8	0.8	0.8	0.4	0.2

Pi-A2 + Pe	0.8	0.6	2.0	0.4	0.4
LSD(P=0.05)	0.21	0.23	0.12	0.35	NS

Table 2. Tuber, stolon and root rot symptom incidences (mean numbers per rep) following solo and combination inoculations of *Phytophthora infestans* (Pi) and *P. erythroseptica* (Pe) in 2002.

Soil Drench Inoculation Treatment	Tuber Pink Rot	Tuber Late Blight	Total Tuber Rot	Total Stolon Rot	Total Root Rot
Control	0	0	0	0	0
Pi, A1 mating type	0	0.6	0.6	0.2	0
Pi, A2 mating type	0	0.1	0.3	0.1	0.2
Pe	0.4	0	0.9	0.5	0.6
Pi-A1 + Pe	0.1	0.2	1.3	0.7	0.5
Pi-A2 + Pe	0.2	0.2	1.3	0.7	0.7
LSD(P=0.05)	0.33	0.55	0.8	0.45	0.40

Table 3. Tuber, stolon and root rot symptom incidences (mean numbers per rep) following solo and combination inoculations of *Phytophthora infestans* (Pi) and *P. erythroseptica* (Pe) in 2004.

Soil Drench Inoculation Treatment	Tuber Pink Rot	Tuber Late Blight	Total Tuber Rot	Total Stolon Rot	Total Root Rot
Control	0	0	0	0	0
Pi, A1 mating type	0	0	0	0.4	0
Pi, A2 mating type	0	0	0.2	0.6	0.6
Pe	0.2	0	0.2	0.6	0.8
Pi-A1 + Pe	0.6	0.6	1.6	0.6	0.2
Pi-A2 + Pe	0.6	0.4	1.0	0.8	0.8

LSD(P=0.05)	0.52	0.44	1.29	NS	0.45
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Isolation and Characterization of Phages Stsc1 and Stsc3 Infecting *Streptomyces scabiei* and their Potential as Biocontrol Agents

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Substantial phage populations exist in the soil with an estimated population of 1.5×10^8 phages/g of soil. Phages represent a potentially important biotic factor affecting bacterial soil communities. Introduction of phage could be used as a control method to limit the population size of plant pathogenic bacteria. The plant pathogen, *Streptomyces scabiei*, is known to colonize and infect potato tubers leading to the development of superficial, raised or deep-pitted brownish lesions that are typical symptoms of common scab. The objective of this study was to isolate and characterize phages infecting *S. scabiei* from soil and to evaluate their potential as biological agents. Phages infecting *Streptomyces scabiei* were isolated from 25 soil samples and their genomes were analyzed by comparing the DNA fragment profile obtained from restriction enzymes digestions. Two genetically different phages infecting *S. scabiei* were found and were named Stsc1 and Stsc3. Phages Stsc1 and Stsc3 had a binal symmetry with an icosahedral head and a long striated tail based on observations using electron microscopy. Bacterial host ranges of Stsc1 and Stsc3 was tested and they infected 88% and 75% of the pathogenic *S. scabiei* strains tested, respectively. Infection of *S. scabiei* strains by phages Stsc1 and Stsc3 was evaluated on solid medium at pH 5.5 to 7.5 and at temperatures varying from 15°C to 30°C. The potential of the phages to be used as biocontrol agents against the pathogen was assessed on radish seedlings *in vitro*.

Development of RT-PCR and Real-Time Quantitative RT-PCR Procedures for Detection and Identification of Potato Virus M

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Potato virus M (PVM, a *Carlavirus*) is considered as one of the common potato viruses distributed worldwide and ELISA is widely employed presently for the detection of this virus in potato samples (tubers, leaves and sprouts). There is a need to introduce molecular diagnosis methods *e.g.* reverse transcription polymerase chain reaction (RT-PCR), restriction fragment length polymorphism (RFLP), DNA array and real-time quantitative RT-PCR (qRT-PCR), to detect PVM in potato samples with a high degree of sensitivity and specificity considering the developments in molecular diagnosis technologies and their application for the detection and identification of several other important potato viruses in recent years. Furthermore, the advantage of quantitative detection of qRT-PCR can be important to determine the quantity of PVM RNA in infected potato samples.

The evaluation of two molecular diagnosis methods, RT-PCR and Cybr Green based qRT-PCR for detecting PVM in potato tissues is described in this report. Based on the analysis of nucleotide sequences of known PVM isolates oligonucleotide primers were designed and evaluated in RT-PCR and qRT-PCR for the amplification of PVM RNA in various potato tissues. The primers developed in this study only amplified PVM RNA in RT-PCR, but not other viruses including potato viruses S, X, Y, potato leafroll virus, potato latent virus, potato mop-top virus and potato isolate of tobacco rattle virus. These primers have a wide range of annealing temperatures (53°C ~ 63°C) that made it simple to include these primers in multiplex RT-PCR detection. Reliable detection of PVM RNA by RT-PCR was achieved in various composite sample sizes (leaves/sprouts: 1/400; dormant tubers: 1/200) and PVM RNAs were detectable by qRT-PCR in all composite samples ranging from pure infected tissue to 1/800 composition. PVM RNA concentration and even copy numbers of PVM RNA molecules can be determined in qRT-PCR test by using a standard curve. Digestion of PCR amplicons with restriction endonuclease *MscI* was demonstrated as a rapid, easy and reliable confirmation method for RT-PCR detection.

Current Research Concerning the Management of Potato Pink Rot, Caused by *Phytophthora Erythroseptica*

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Pink rot, caused by *Phytophthora erythroseptica* Pethyb., is an important disease of potatoes in many growing regions. Pink rot can cause pre- and post-harvest yield losses for potato growers, particularly during growing seasons with excessive precipitation. All underground potato tissues (roots, stolons, tubers, basal stems) may be affected, but diseased tubers are most commonly observed. Tubers are usually infected at the stem end via diseased stolons. However, when enough moisture is available, zoospores (swimming spores) can infect tubers through buds or lenticels.

Infected tubers become spongy and a dark line separates diseased from healthy tissue. Lenticels in the infected areas of the tuber often become blackened. When infected tubers are cut open, diseased tissues appear creamy and exude a clear fluid when squeezed. Infected tuber tissues on cut surfaces turn pink when exposed to the air for 20-30 minutes.

Cultural management of pink rot

Most soils in potato-growing regions of North America contain oospores (long-lived survival spores) of *P. erythroseptica*. However, infected seed tubers can also spread new strains of the pathogen between regions, thus planting disease-free seed is an important first step in disease control. There is some evidence that crop rotation can reduce levels of inoculum in the soil. Pink rot is most severe under wet soil conditions, so providing good soil drainage, delaying harvest when soils are wet and warm (>18°C), and avoiding harvest of wet areas are good management practices to follow. In addition, minimizing tuber injury at harvest and grading out diseased tubers prior to storage will reduce storage rot. Tubers from 'questionable' fields should be stored last and storage conditions should be monitored carefully.

Cultivar susceptibility to pink rot

In recent years, we have screened several potato cultivars for resistance to pink rot using tissue culture microplantlets or harvested tubers. The microplantlet method allows an examination of the response of non-tuber tissues to disease (plant wilting; browning and death of plant stems and leaves). This method may also have some predictive value for

estimating the potential of tubers to become infected, since most tubers in the field are infected via diseased stolons. Our studies have shown that although no commonly-grown potato cultivars are immune to pink rot, varying degrees of disease resistance can be found in different cultivars. Of 20 cultivars examined, Goldrush, AC Novachip, Yukon Gold, Shepody, and Russet Norkotah were the most susceptible to disease, while Ranger Russet, Snowden, Butte, and Russet Burbank were the least susceptible. In general, microplantlets of potato cultivars with late-season field maturity were more resistant to disease than those with early or mid-season maturity. In most cases, the ranking of cultivars for disease susceptibility following direct infection of harvested tubers was similar to the microplantlet results. However, some differences were observed. For example, Yukon Gold and Goldrush were less susceptible to direct infection of tubers than they were to infection of non-tuber tissues in the microplantlet screen. Both tuber and non-tuber tissues need to be evaluated to best determine the resistance of a cultivar to pink rot. Growers have the option of growing less susceptible cultivars in fields with drainage problems or a history of pink rot.

Pink rot and late blight

Recent studies at the Crops and Livestock Research Centre have examined the interaction of *P.infestans* (late blight) and *P. erythroseptica* (pink rot) in potato storage, greenhouse, and field trials. The two pathogens can commonly be found co-infecting potato tubers. Early results indicate that tuber rot severity is significantly increased in the presence of both pathogens. Successful management of late blight may also aid in the management of pink rot.

Mefenoxam sensitivity of *P. erythroseptica*

Chemical control of pink rot relies on the use of the systemic fungicide mefenoxam (Ridomil® Gold). In recent years, the development of mefenoxam-resistant strains of *P. erythroseptica* in the U.S., particularly in Maine and Idaho, has caused concern. Surveys conducted from 1999 to 2001 in PEI indicate that populations of the pathogen in PEI are still sensitive to mefenoxam and Ridomil Gold would be an effective product to use as part of a pink rot management program. More extensive surveys of pathogen populations in Canadian potato-growing regions are needed to determine the prevalence of mefenoxam-resistant strains in this country.

Use of Ridomil Gold to control pink rot

Between 2000 and 2004, field trials were conducted at locations across Canada (Alberta, Manitoba, Ontario, New Brunswick and Prince Edward Island) to compare foliar with in-furrow applications of Ridomil Gold for control of pink rot in harvested tubers. Comparisons were made between a standard Ridomil Gold/Bravo foliar program and Ridomil Gold 480EC applied as a 15 cm band in-furrow at planting. Cultivars grown included Russet Burbank, Yukon Gold and Shepody. Tubers were harvested from field plots in September and then inoculated with either mefenoxam-sensitive or mefenoxam-resistant isolates of *P.*

erythroseptica.

Foliar field applications of mefenoxam reduced disease in tubers in 2000, but were largely ineffective in 2001 and 2002 (Table 1). Inconsistency in the ability of foliar applications of mefenoxam to control pink rot may be due to poorly timed spray applications, differences in cultivar response, and environmental factors. For example, drought conditions in 2001 likely restricted the movement of mefenoxam from the leaves to the tubers resulting in poor disease control.

By contrast, in-furrow applications of Ridomil Gold have provided excellent suppression of pink rot in most years of the study (Table 1). However, some variation in efficacy has still been noted among regions, so that in-furrow applications of mefenoxam in eastern Canada tend to provide better disease suppression than identical applications in western Canada. Whether soil type, climatic conditions or other factors are responsible for this variation requires further investigation. Certainly, the mobility of mefenoxam (or metalaxyl) in soil depends on the pH, the organic matter, and the clay content of the soil, as well as rainfall amounts following soil application, and a potential for leaching of the fungicide out of the root zone does exist. Another possibility is that mefenoxam is bound to the clay particles making it unavailable to the plant. Nevertheless, early season applications of mefenoxam may address some of the factors responsible for inconsistent control of tuber rot with foliar applications and allow for better positioning of fungicidal compounds near to or within underground plant tissues. Since most tuber infections originate at the stem end following stolon infection, applications of mefenoxam at planting may be more appropriate than foliar applications to circumvent tuber infection by this pathway.

Applications of mefenoxam (foliar or in-furrow) did not control pink rot in tubers inoculated with mefenoxam-resistant isolates of *P. erythroseptica*. This confirms the importance of fungicide resistance management. Research is required to examine the impact of the in-furrow application method on the development of mefenoxam-resistant pathogen populations. Continued monitoring of the mefenoxam resistance of populations of *P. erythroseptica* will ensure that mefenoxam is applied where mefenoxam-sensitive populations of the pathogen occur.

Table 1. Percent (%) of surface rot in tubers from various Canadian field trials inoculated with a mefenoxam-sensitive isolate of *Phytophthora erythroseptica*.

Treatment	2000		2001				2002			
	NB	PE	MB	ON	NB	PE	MB	ON	NB	PE
Bravo Check	86	50	40	65	49	71	33	51	55	50
Ridomil Gold/Bravo Foliar	20	33	43	64	43	72	28	42	53	26

Reverse Transcription-Loop Mediated Isothermal Amplification of DNA for Detection of Potato Viruses

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Development of sensitive and efficient pathogen diagnostic techniques which are cost effective and environmentally friendly is of interest to the potato industry locally, nationally and globally. Recently, a nucleic acid-based technique termed “Loop mediated isothermal amplification of DNA (LAMP)” has been invented (Notomi et al., 2000, *Nucleic Acids Res.* 28:e63.). Using *Potato virus Y* (PVY) as a model system, the efficacy of LAMP for detection of potato viruses has been assessed. Four primers matching a total of six sequences of the coat protein (CP) gene of PVY were designed in such a way that a loop could be formed and elongated during DNA amplification. The LAMP reaction was optimized by adjusting the concentrations of various components. The effects of fragment length of the target DNA on LAMP were also investigated. Two-step and one-step RT-LAMPs were performed using RNA extract of various PVY cultures, and the results were correlated with two-step reverse transcription-polymerase chain reaction (RT-PCR) detection of PVY. Further, the turbidity caused by precipitation of magnesium pyrophosphate formed in a positive RT-LAMP reaction was used to measure the amplification using a time-saving spectrophotometric method. The preliminary research indicates that this method has excellent potentials for diagnosis of potato viruses.

Occurrence of *Phytophthora infestans* on Hairy Nightshade in Maine: Disease Implications of Isolates from Divergent Hosts and Genotypes

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Occurrence of *Phytophthora infestans* on hairy nightshade (*Solanum sarrachoides*) was detected during the 2004 cropping season in Maine. Allozyme, mating type, epidemic components and potential for sexual reproduction of isolates from diverse hosts and genotypes were assessed in laboratory and growth chamber assays. The inoculum contribution of isolates derived from hairy nightshade was evaluated in cross-inoculation studies. Isolates from hairy nightshade and potato were 100/111/122 genotype and A2 mating type based on allozyme analysis. In cross-inoculation studies, when inoculum was derived from hairy nightshade, the infection frequency of potato and nightshade leaves was 67 and 50%, respectively. When the inoculum source was infected potato, the infection frequency was 83 and 63%, respectively. Progress of late blight incited by isolates from hairy nightshade and potato was similar, and ranged from 54 to 87% after 12 days of incubation. Lesion growth from divergent genotypes was significantly different at various incubation temperatures ($P < .05$). The highest disease severity was detected on 100/111/122 genotype. In-vitro oospore production varied among crosses of isolates from diverse hosts and genotypes, and the percentage of crosses resulting in oospore production ranged from 0 to 67%. These studies suggest that hairy nightshade is a potential source of late blight inoculum, and disease components can be impacted by genotype and host diversity.

Suppression of Early Blight (*in vivo*) and Germination of *Alternaria* spp. Conidia (*in vitro*) with Azoxystrobin

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Experimental potato field trials were established in New Annan, Prince Edward Island, to assess the efficacy of strobilurin fungicides (Quadris® and Headline®) to suppress *Alternaria solani*, causal agent of potato early blight. At the end of the growing season, these field trials were rated according to the Horsfall-Barrett scale and an obvious fungicide treatment effect was found. The incidence and severity of early blight was low in plots treated with strobilurin fungicides compared to untreated plots where disease was more severe. In addition to the foliage ratings, isolates of *A. solani* as well as *Alternaria alternata*, another potato leaf spot pathogen, were collected from the experimental trials and several other potato fields for subsequent laboratory work. An *in vitro* spore germination assay was used to measure the sensitivity of these isolates to azoxystrobin, the active ingredient in Quadris® fungicide. The effective concentration that inhibited spore germination by 50% (EC₅₀) was determined for each isolate. EC₅₀ values ranged from 0.003 to 0.014 parts per million (ppm) for *A. solani* while the values for *A. alternata* ranged from 0.001 to 0.023 ppm. These results suggest that the isolates tested are sensitive to azoxystrobin; no indication of resistance was observed. This sensitivity is likely due to the limited exposure of these two pathogens (*A. solani* and *A. alternata*) to strobilurin chemistry in Prince Edward Island potato fields.

Factors Controlling the Rate and Partitioning of Gaseous Nitrogen Losses from Denitrification

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Nitrous oxide (N₂O) accounts for the majority of Canadian agricultural greenhouse gas emissions and, in humid regions, is associated primarily with the process of denitrification. An understanding of the controls of the denitrification process is required to design management strategies to reduce N₂O emissions from agricultural soils. The purpose of this study was to identify the key factors controlling the rate and partitioning (N₂O:N₂) of gaseous N losses from denitrification. Denitrification (N₂O + N₂) and N₂O emissions were measured on repacked soil cores using the acetylene inhibition technique. Three laboratory incubations were carried out to investigate the effect of additions of nitrate as KNO₃ and carbon as glucose, and their interaction on both total denitrification and the partitioning (N₂O:N₂) of gaseous N losses from denitrification. In the first incubation, NO₃-N addition had no significant effect on denitrification, but increased N₂O emissions. In the second incubation, the addition of a carbon source resulted in increased N₂O emissions with increasing nitrate concentration. Denitrification, however, showed no response to nitrate addition, even in the presence of carbon. In the third incubation, NO₃-N addition had no significant effect on denitrification or N₂O emissions and the addition of a carbon source generally increased N₂O emissions and significantly increased denitrification. While the 62.5 mg C kg⁻¹ soil treatment addition emitted 0.73 mg N₂O-N kg⁻¹ soil as N₂O emissions and 1.02 mg N₂O-N kg⁻¹ soil from denitrification, the 500 mg C kg⁻¹ soil treatment addition emitted 4.93 mg N₂O-N kg⁻¹ soil as N₂O emissions and 6.80 mg N₂O-N kg⁻¹ soil from denitrification. These results point to the critical role of carbon availability as a key factor controlling the rate and partitioning (N₂O:N₂) of gaseous N losses from denitrification. The native NO₃⁻ content in the soil was sufficient to support increased denitrification in response to carbon addition and was not further stimulated by the addition of NO₃⁻. The amount of N₂O released from denitrification (N₂O:N₂) was significantly increased following the addition of nitrate. This result highlights the important role of competition between terminal electron acceptors (NO₃⁻, NO₂⁻, N₂O) in determining the final productions of the denitrification process, specifically the relative amounts of N₂O and N₂ released from the soil.

Genetic Variation Among PLRV Isolates in Prince Edward Island and the Detection of PLRV in Potatoes

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Potato leafroll virus (PLRV)(genus *Polerovirus*, family *Luteoviridae*), a major potato (*Solanum tuberosum* L.) pathogen, is mainly transmitted through infected seed tubers and by aphids. Plants developed from PLRV infected seed tubers show a characteristic leaf-roll symptom, and the number of stems per plant, number and size of marketable tubers can be significantly reduced. Yield reduction due to the use of PLRV infected seed tubers has been reported for many commercial potato varieties, and the detection of PLRV in seed tubers has been an integral part of seed potato certification programs worldwide. TAS-ELISA is the dominant method currently in use for screening potato samples for PLRV. Nucleic acid hybridization (NCH) and RT-PCR have also evaluated for detecting PLRV in various potato tissues. Inconsistency between results obtained by ELISA and by molecular diagnostic method (NCH or RT-PCR) has been reported and the cause was generally considered as low virus titre in potato tubers and low sensitivity of ELISA. In this report, the identification and characterization of eleven PLRV isolates from an experimental farm in Prince Edward Island (PEI) are described. Different diagnosis methods were employed for the testing of the same tuber samples for the presence of PLRV RNAs and the nucleotide and amino acid sequences of the coat protein (CP) gene of different PLRV isolates were analysed to determine the genetic variability among PLRV isolates and mutation in PLRV CP.

Eleven PEI PLRV isolates were evaluated and one isolate from the virus collection at CFIA Charlottetown lab was included in the study as the reference. These isolates were maintained in potato plants (Shepody). Tubers harvested from PLRV infected plants were initially screened by RT-PCR and three PLRV positive tubers from three plants of each isolate were chosen for further evaluation. These tubers (in dormant stage) were tested by TAS-ELISA, quantitative dot-blot hybridization (qDBH) using a DIG-cRNA probe specific to PLRV CP gene and RT-PCR targeting CP gene sequence. RFLP was conducted to verify all PCR amplicons. Most of these tubers were confirmed to be negative to PLRV in TAS-ELISA. All tested tubers were positive to PLRV in qDBH and RT-PCR tests and relative PLRV RNA concentration was determined by qDBH. PCR amplicons were produced from all 11 PEI PLRV isolates and the reference isolate and nucleotide sequences of the PCR products were then determined by automated cycle sequencing. A moderate level of genetic variation either based on nucleotide or amino acid sequence of the CP gene was revealed. But, amino acid replacement mutations occurred in the CP of tested PLRV isolates may not

be the cause that they were undetectable in potato tubers by ELISA. Phylogenetic analysis of sequences of these isolates was conducted using Align Plu 4.0 and Blast was performed to identify related sequences available from various databases. A phylogenetic tree was developed from the data by neighbour-joining algorithm (bootstrap 50% value). PLRV isolates from all different regions of the world were divided into several isolate groups.

Evaluation and Selection of Simple, Rapid and Cost-Effective RNA Extraction Procedures for Detecting Potato Viruses

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Several diagnostic methods targeting viral RNAs, such as nucleic acid hybridization, DNA array, RT-PCR and real-time quantitative RT-PCR (qRT-PCR) have been developed and demonstrated to have many advantages over those widely used serological and biological diagnostic methods such as ELISA, Western blot and bioassay, *e.g.* sensitivity, specificity, simplicity and speed. The first and critical step for detecting RNA viruses of potato is to extract high quality total RNAs or viral RNAs from infected potato tissues. Conventionally homogenized potato tissue or tissue sap is extracted with buffer or water saturated phenol followed by the removal of phenol with chloroform and precipitation of total nucleic acids (DNAs and RNAs) by ethanol at low temperature. This method is time consuming and requires the repeated transfer of solutions and repeated centrifugation steps. This concern, particularly in the situation where a large number of samples are to be extracted. Furthermore, an incomplete removal of phenol in the final nucleic acid preparation will possibly inhibit enzymatic reactions down stream, particularly polymerase chain reaction in RT-PCR and qRT-PCR.

In recent years, great effort has been made to modify and improve RNA (total RNAs, mRNAs) extraction procedures. Many biotech firms have developed RNA extraction kits which are now available in the market. Evaluation and/or validation of new methods and commercial kits for potato virus testing are necessary. In this report the evaluation of several new/modified RNA extraction methods and/or commercial kits for total plant RNA extraction is described. The yield and quality of RNAs obtained were examined before the RNAs were used in RT-PCR and qRT-PCR tests. High efficiency of amplification in qRT-PCR was observed from those RNAs extracted by using Plant Concert RNA Reagent, RNeasy mini kit, Promega SV RNA kit and Tri-Reagent solution from the same potato sample infected with a virus (PVY^{NTN} or PVM). Several other aspects of RNA extraction procedures, such as cost per extraction, overall time needed and tube changes/solution transfers, were also evaluated. The possibility of using the RNA extraction method or commercial RNA extraction kit for an automated or semi-automated total plant RNA extraction was also assessed.

Nitrogen Influx Kinetic Parameters of Potato Roots

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Potatoes (*Solanum tuberosum* L.) are a relatively shallow rooted crop, and often are provided with high rates of fertilizer N. The shallow root system and high N application rates increase the risk of nitrate leaching to groundwater and of nitrous oxide emissions. One strategy to reduce environmental losses of nitrogen is to improve the efficiency of the potato crop with respect to nitrate uptake. Knowledge of the N flux characteristics of potato roots is important in better understanding of potato nitrogen uptake efficiency mechanisms and efficient use of N fertilizers. Estimates of the kinetic uptake parameters for NO_3^- , as the main source of plant N uptake, have been reported for several crops, but not for potato. These kinetic parameters include I_{max} (maximum uptake rate), K_m (Michaelis-Menten constant) and C_{min} (minimum concentration at which uptake can occur). In addition, there is no information on the degree of variation in nitrate influx kinetic parameters among potato cultivars. Therefore, the objective of this study was to measure and compare the nitrate influx kinetic parameters among potato cultivars at different plant growth stages.

A hydroponic experiment with five potato cultivars (Atlantic, Chieftain, Red Pontiac, Russet Norkotah and Shepody) and three plant growth stages (20, 27 and 32 days after transplanting of plantlets from tissue culture to solution culture (DAT)) was conducted in a growth room. At each growth stage, depletion experiments (depletion of NO_3^- from solution by roots during the time) were conducted with three replications of five potato cultivars using a depletion method similar to Claassen and Barber (1974). The plants were harvested after depletion experiments and roots were scanned, analyzed by WinRHIZO Version 2002C PRO software and root length (RL) was calculated. The nitrate concentration of solution samples was determined colorimetrically using a Technicon TRAACS 800 auto-analyzer with 0.02 mg L^{-1} NO_3^- -N detection limit. A depletion curve, plot of nitrate concentration in solution against sampling time, was developed and the kinetic uptake parameters were calculated as describe by Claassen and Barber (1974).

The I_{max} values were between 0.79 to 7.62 with average of 2.44 $\text{pmol cm}^{-1} \text{s}^{-1}$. This is in the range of herbaceous plants reported by other authors at almost comparable plant age range, for example 0.20 to 2.50 $\text{pmol cm}^{-1} \text{s}^{-1}$ for lettuce (Steingrobe and Schenk, 1994) and 0.3 and 0.6 $\text{pmol cm}^{-1} \text{s}^{-1}$ for spinach and kohlrabi, respectively (Steingrobe and Schenk, 1991). The maximum nitrate inflow was below 100 $\mu\text{M NO}_3^-$ at early growing stages and below 50 $\mu\text{M NO}_3^-$ thereafter. The measured I_{max} range was lower compared to ranges reported for cereals and grasses (Barber, 1995) which may imply that potato is less effective

in N uptake compared to cereal and consequently requires high N rates and has a higher risk of nitrate leaching. There were significant differences in nitrate I_{max} among cultivars at 20 DAT after which the differences were limited. Russet Norkotah and Chieftain had significantly higher I_{max} than other potato cultivars at all three growth stages. It could be related to their lower root:shoot ratio ($m\ g^{-1}$) compared to the other cultivars. The I_{max} decreased with plant age (from about $5.10\ pmol\ cm^{-1}\ s^{-1}$ at 20 DAT to $0.98\ pmol\ cm^{-1}\ s^{-1}$ at 32 DAT).

Calculated values for K_m ranged from 11.21 to 17.24 μM and for C_{min} ranged from 0.94 to 2.01 μM with the highest values were measured for Red Pontiac. This implies that Red Pontiac could be more sensitive to N deficiency than other cultivars. There was no significant effect of plant age on K_m and C_{min} . Cultivars differed primarily in I_{max} values rather than K_m and C_{min} values.

The results suggest that Chieftain has the capacity to absorb more NO_3^- in each unit of their root length than other cultivars in the same soil. Russet Norkotah also has a high uptake per unit root length, but this characteristic is of limited value due to the limited root system for this cultivar. Therefore, one of the most important uptake efficiency components for potato crop is maximum N inflow rate, which showed significance differences among the investigated cultivars. Screening based on the kinetic parameters of N uptake by potato roots might be used to select more N uptake efficient cultivars.

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Microclimatic Parameters and Potential for Late Blight Development in Irrigated Potato in Maine

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Application of irrigation water can improve potato growth and tuber yield in years of deficit rainfall. However, its effect on microclimate and potential for potato late blight development are not fully understood. The effect of sprinkler, sub-surface drip, and surface drip irrigation treatments on microclimatic parameters indicative of late blight susceptibility were assessed on a Russet Burbank potato in 2003 and 2004. Canopy temperature, relative humidity (RH), rainfall, leaf wetness and soil temperatures were recorded with a data-logger. Based on microclimatic data recorded within the plant canopy, the potential for late blight was assessed using a simulation model. Average canopy temperatures were lowest with sprinkler and surface drip irrigation in 2003 but not in wetter 2004 cropping season. Cumulative time with RH above 90% and temperature less than 22 C was consistently greater in sprinkler and surface drip treatments in 2003, but no treatment effect was evident in 2004. The Simulated area under disease progress curves was slightly higher in irrigated than non-irrigated treatments. These results indicate higher potential for late blight when potatoes are irrigated, however, this increase is minimized by applying irrigation water through sub-surface drip systems.

Production of Fermentable Sugars from Potato Waste for the Use in Bioethanol Production

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Bioethanol produced from potato waste has a large potential market. If federal government regulations are adopted in light of the Kyoto accord, the mandated blending of bioethanol with traditional gasoline in amounts up to 10% will result in requirements of large quantities of bioethanol in Canada. In addition to fuel bioethanol there are also a number of markets for pure 99% industrial ethanol in the form of vodka or for other industrial uses. In the potato belt region of New Brunswick there is a significant amount of potato waste produced from processing and in the form of culls. The utilization of ethanol production as a waste disposal method and the production of a value added product is of benefit to both the potato industry and the environment. The fermentable sugar values of steam peel waste from the Grand Falls McCain plant, CANUSA waste (white water and steam peel) and of local cull potatoes, was determined during a period from September 2003-March 2004. The steam peel was found to be $11.6\pm 3.2\%$ dry matter, $8.5\pm 4.5\%$ starch and $3.4\pm 4.7\%$ fermentable sugars (maltose and glucose). Culls were $10.2\pm 0.3\%$ dry matter, $9.7\pm 1.4\%$ starch and $0.5\pm 1.3\%$ fermentable sugars (maltose and glucose). CANUSA waste was approximately 50% dry matter, 64% starch. A number of the batches of steam peel and culls were fermented to determine fermentability and ethanol production. All sources of waste were considered to be fermentable and steam peel produced on average a potato beer containing 1% ethanol, CANUSA waste 3% and Culls 0.5% (note: low ethanol percentage are due to the high percentage of water in the potatoes and in the fermented potato mixture). The nutrient value of steam peel, culls and CANUSA stillage was similar in value to dry distillers grains from corn. This means that there would be no negative effects on the nutrient value of the stillage or dry distiller grains from blending potato waste with other feedstocks in the ethanol fermentation process. A multiple feedstock process composed of potato waste and grain (barley or corn) is probably the most viable option for the potato region of New Brunswick. The amount of available processing waste combined with the lower than expected starch values means that the current volume of processing waste can produce 4-5 million litres (Based on a rough estimate of 44000 tonnes of processing waste per year from all the processors in the region). The remaining ethanol output volume in a small-scale 10-15 million litre plant could easily be obtained from local grain.