PEPS (PGPR-Enhanced Phytoremediation Systems): The Chemistry and Biology Behind Successful Phytoremediation of Petroleum and Salt Impacted Soils

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Partners: Cenovus; ConocoPhillips; Lone Pine; Baytex; TransEuro; Shell; Devon; Tundra; Enbridge; Seaway Energy Services; MWH; Stantec; SLR; NSERC

The Phytoremediation Process

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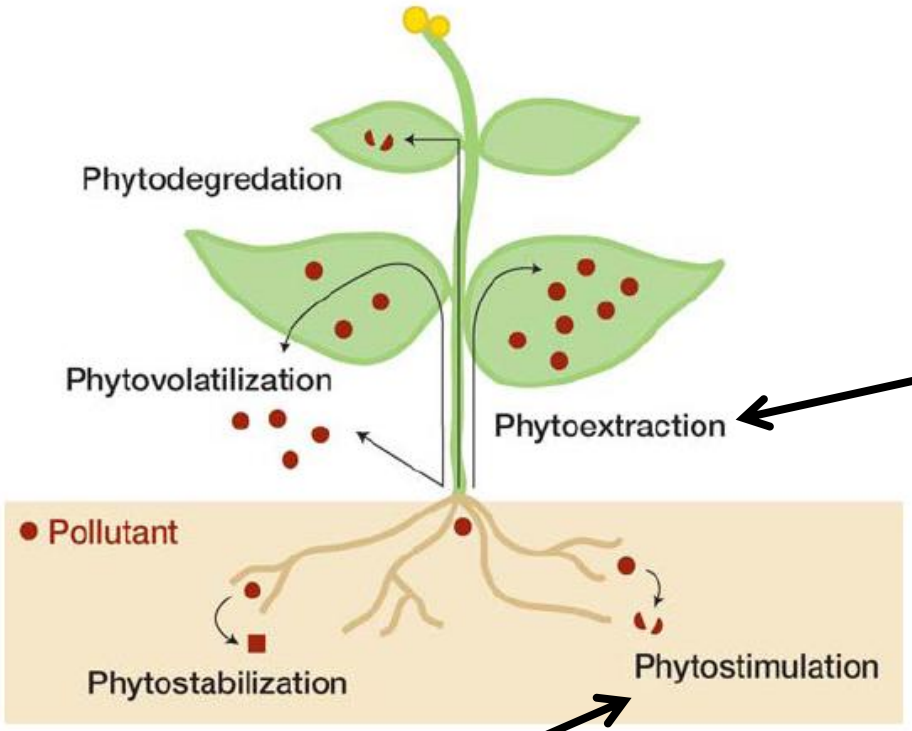
Phytoremediation



Salt

- Volatilization
- Phytodegradation
- Chelation/compartment in leaves

- Translocation: root symplast→ xylem
- Chelation/compartment in roots
- Plant uptake soil→root
- Rhizosphere Processes
- Bioavailability particle→water

Rhizodegradation - PHC

Advantages of Phytoremediation

1. Soil quality improved
2. Driven by solar energy - suitable to most regions and climates
3. **Cost effective**
4. **Plants** provide **sufficient biomass** for rapid remediation; **promote high rhizosphere activity**
5. Reasonable time frames - 2 to 3 years
6. Can be used effectively at **remote sites**
7. Greenhouse gas storage: **6 tonnes per ha per year**
8. Effective for remediation of PHC and salt – relevant to the energy industry

Development, Proof and Full Scale Application of PGPR Enhanced Phytoremediation Systems (PEPS)

Over 13 years of research with full-scale field remediations at each stage of development and application

1. **PHC:** sites in AB, BC, QC, MB, NWT and ON (2004-12)
2. **Salt:** sites in SK, AB and NWT (2007-12)

Performing full scale remediations for > 7 yrs

PEPS currently successfully operating at > 30 sites

> 10 sites completed

The key to phytoremediation success -
Transfer of the science from the lab to the field

1. Strategies for aggressive plant growth in impacted and poor quality soils at full scale sites
   – PEPS Deployment by highly trained scientists
2. Monitoring the progress of phytoremediation at each site – Following the chemistry
3. Continuous improvement of our phytoremediation systems through scientific research

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WEBi-Earthmaster-UW Partnership

- Synergistic expertise in contaminated site remediation
- Developed commercial phytoremediation technologies (PEPS)
- > 10 years of research, development and full scale field implementation
- Field proven systems
- Research to continually improve PEPS

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PGPR Enhanced Phytoremediation Systems (PEPS)

Aggressive plant growth strategies leads to remediation

Physical soil treatment: Seed bed preparation

Phytoremediation: Growth of plants **with PGPR**

Monitoring and remediation assessment: Environmental chemistry to follow PEPS from start–to–finish

- PGPR: Plant growth promoting rhizobacteria.
- Prevent the synthesis of stress ethylene.
- PGPR are applied to the grass seeds prior to sowing
  → **NOT Bioaugmentation**
- Effect depth of remediation ~ 0.5 m

"So, what have we been doing for the last year?"

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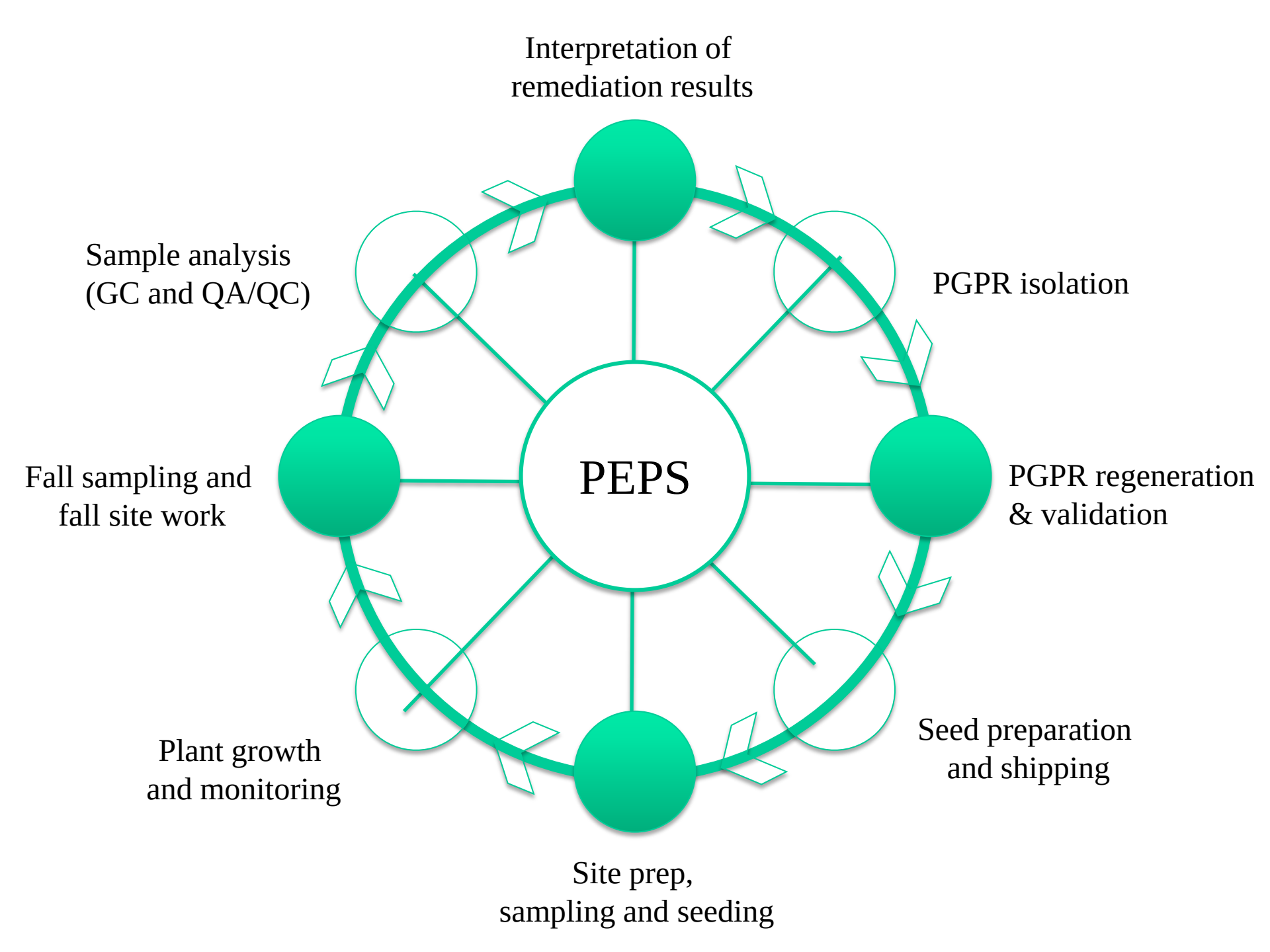
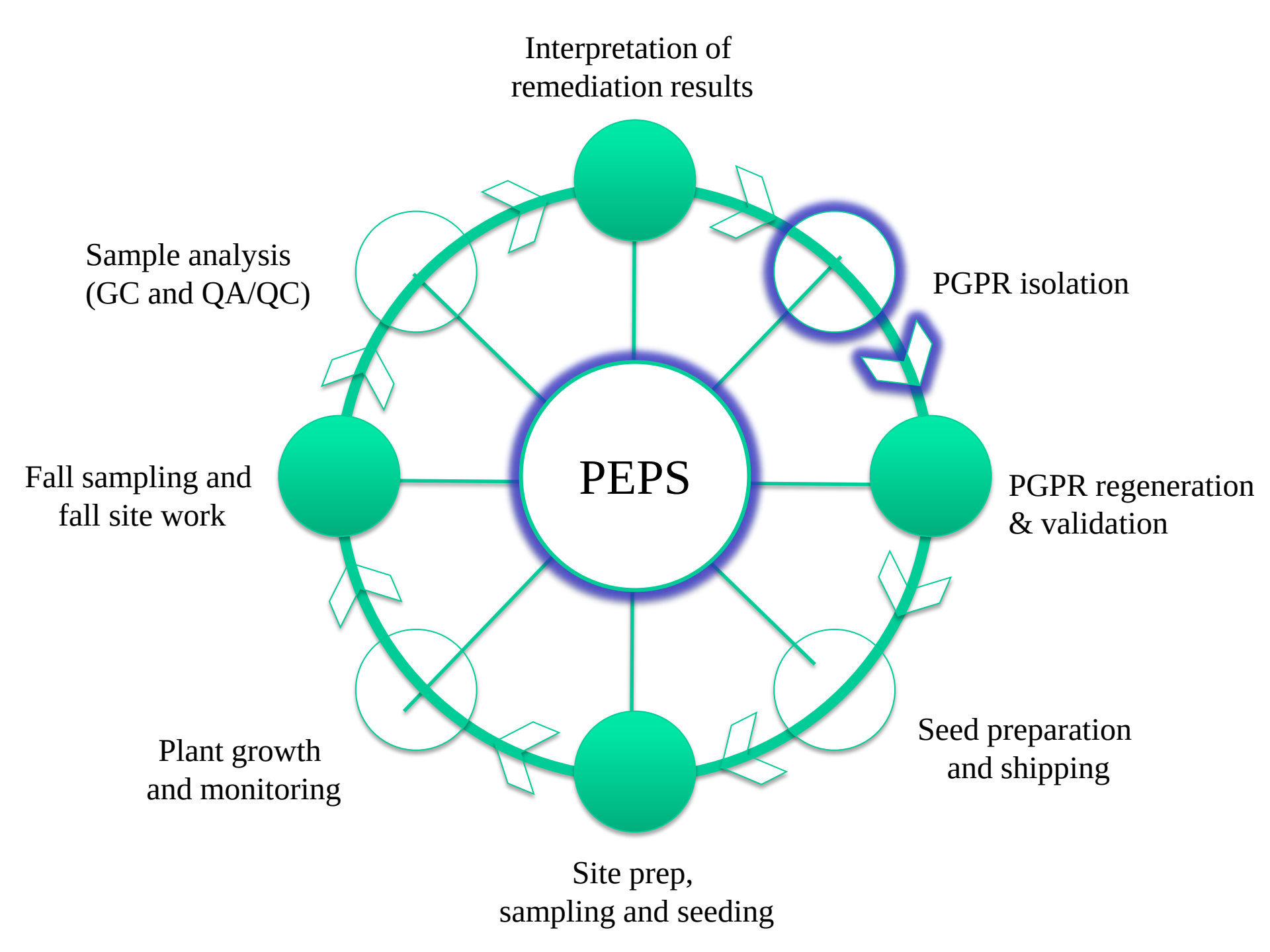

Interpretation of remediation results
Sample analysis (GC and QA/QC)
PGPR isolation
PEPS
Fall sampling and fall site work
PGPR regeneration & validation
Plant growth and monitoring
Seed preparation and shipping
Site prep, sampling and seeding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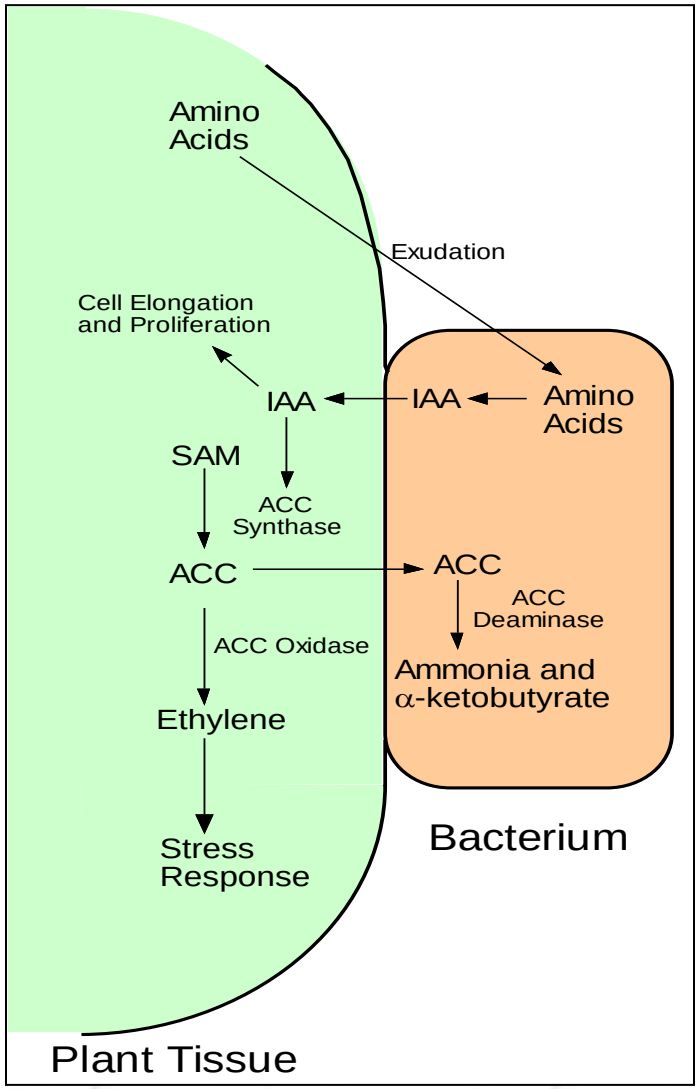
Interaction of a PGPR Containing ACC Deaminase with a Plant Seed or Root



Plant growth promoting rhizobacteria (PGPR)

Natural, non-pathogenic strains of PGPR (usually *Pseudomonads*)

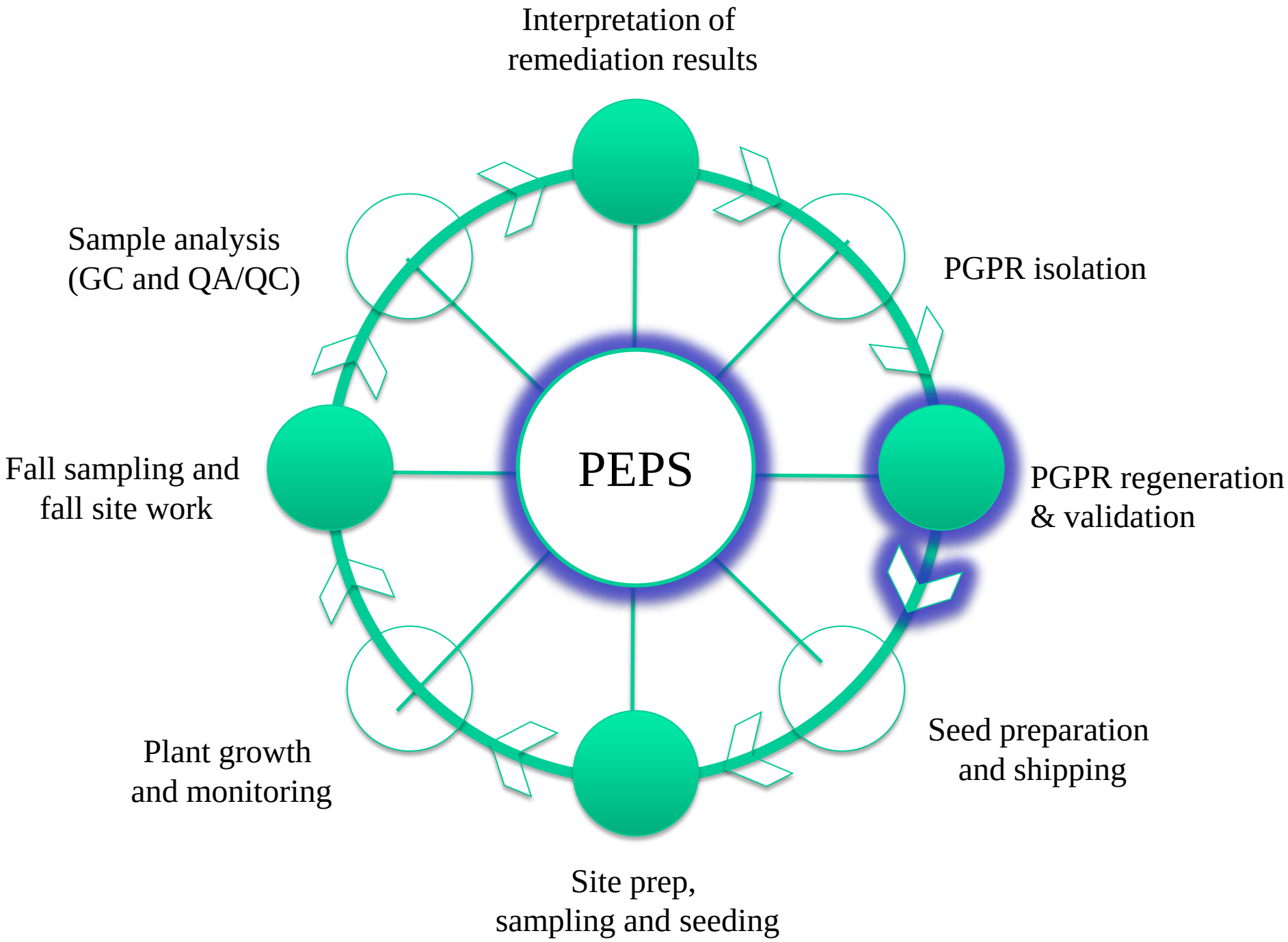
We have isolated PGPRs from ON, AB, SK and the NWT

PGPR are applied to seeds prior to planting

PGPR Isolation

Plant Growth-Promoting Rhizobacteria (PGPR) Isolation

- Have great PGPRs already – assume better strains in environment
- New PGPR continually being isolated
- Successful strains are assayed further by DNA sequencing to identify the bacteria. Only those in Biosafety Level 1 used. All non-GMOs
- Isolated from site rhizosphere soils - environments we work in so they are adapted to impacted soils
- These will be PGPR we will use in the future
- New IRAP funding for this research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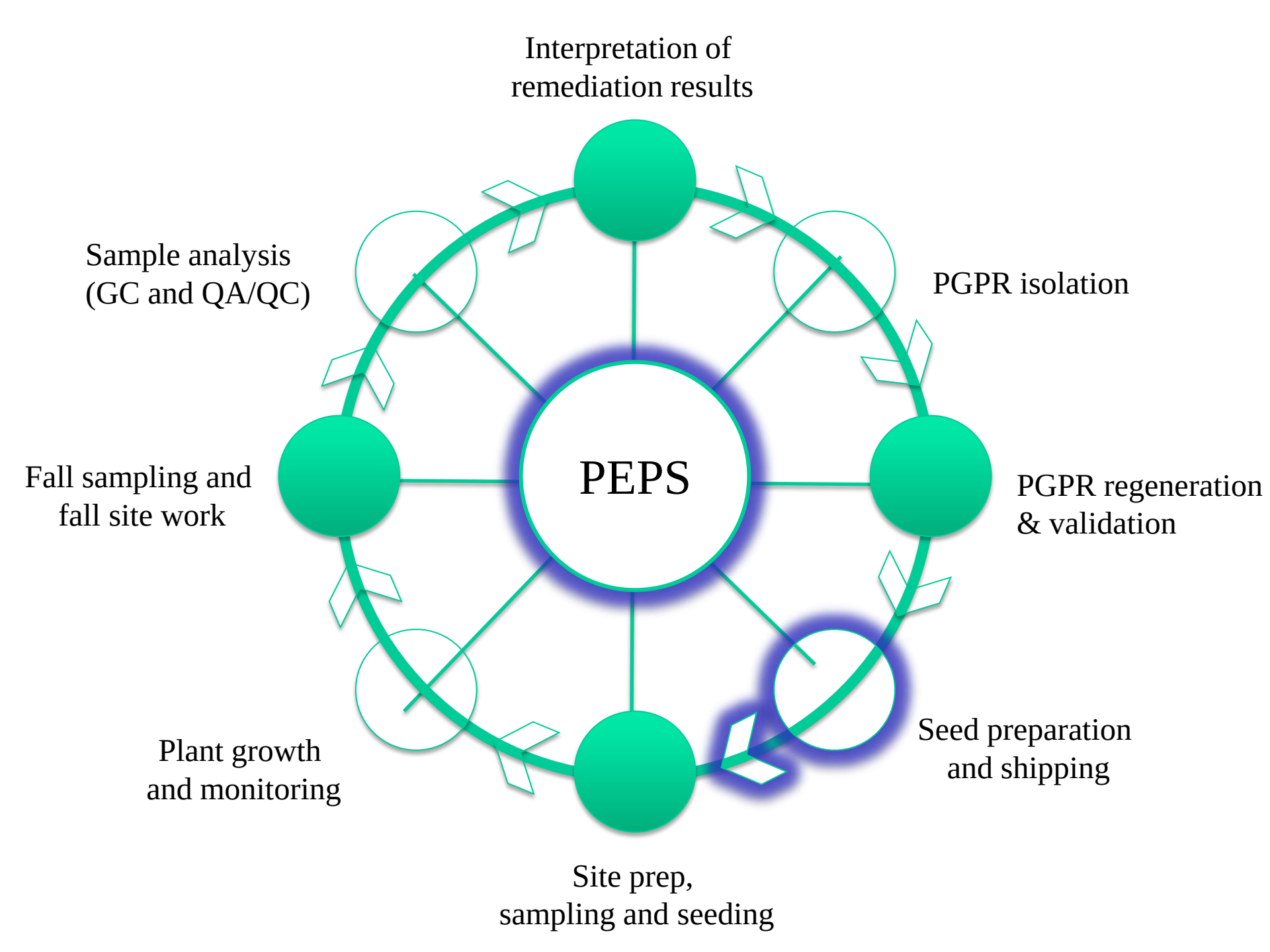
PGPR Regeneration & Validation

Regeneration of proven PGPR isolates for field use

- Currently have > 10 strains of proven PGPR
- Every year before use in PEPS – must confirm they are healthy and retain key biological activities for active plant growth promotion
- Assay for ACC Deaminase
- Assay for Auxin production

PGPR Validation – Assess plant growth enhancement

- Before use in PEPS: plant growth assays to assure PGPRs perform properly
- PGPRs now ready for use in PEPS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Seed Treating & Shipping

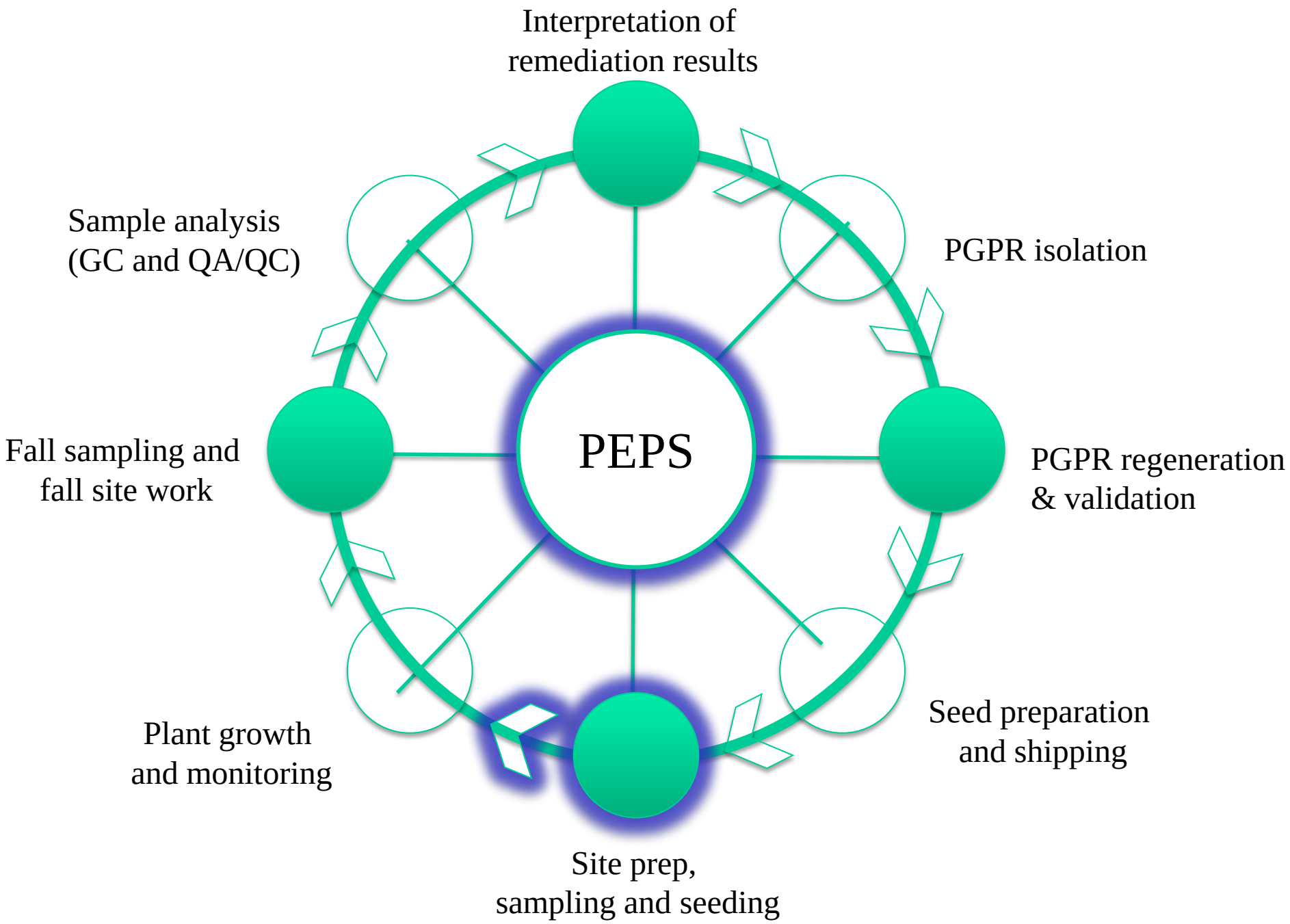
- Treat seeds with proven and regenerated PGPR for field deployment
- Only proven grass and cereal species are used
- Mechanical seed treater efficiently and evenly coats the seeds
- Dried seed rapidly produced

PGPR Seed Treatment QA/QC

Aliquots of PGPR-treated seeds assayed for plant growth enhancement to assure successful PGPR application

Seed Treating & Shipping

- Treated seeds shipped to sites after QA/QC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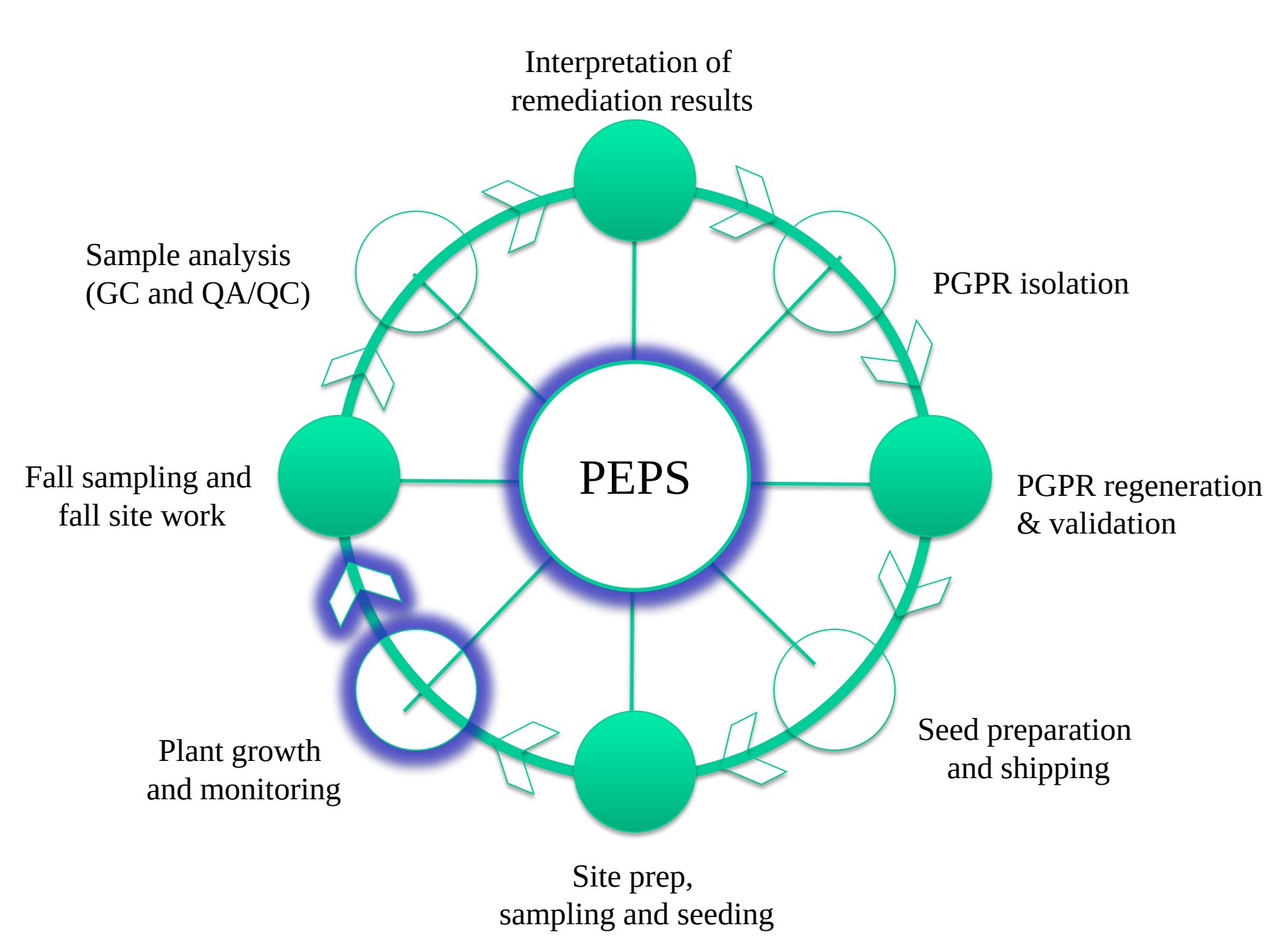
Disking/Tilling to prepare seed bed

Application of appropriate fertilizer

Sample soil for beginning of season PHC and/or Salt levels

Sow PGPR-treated seed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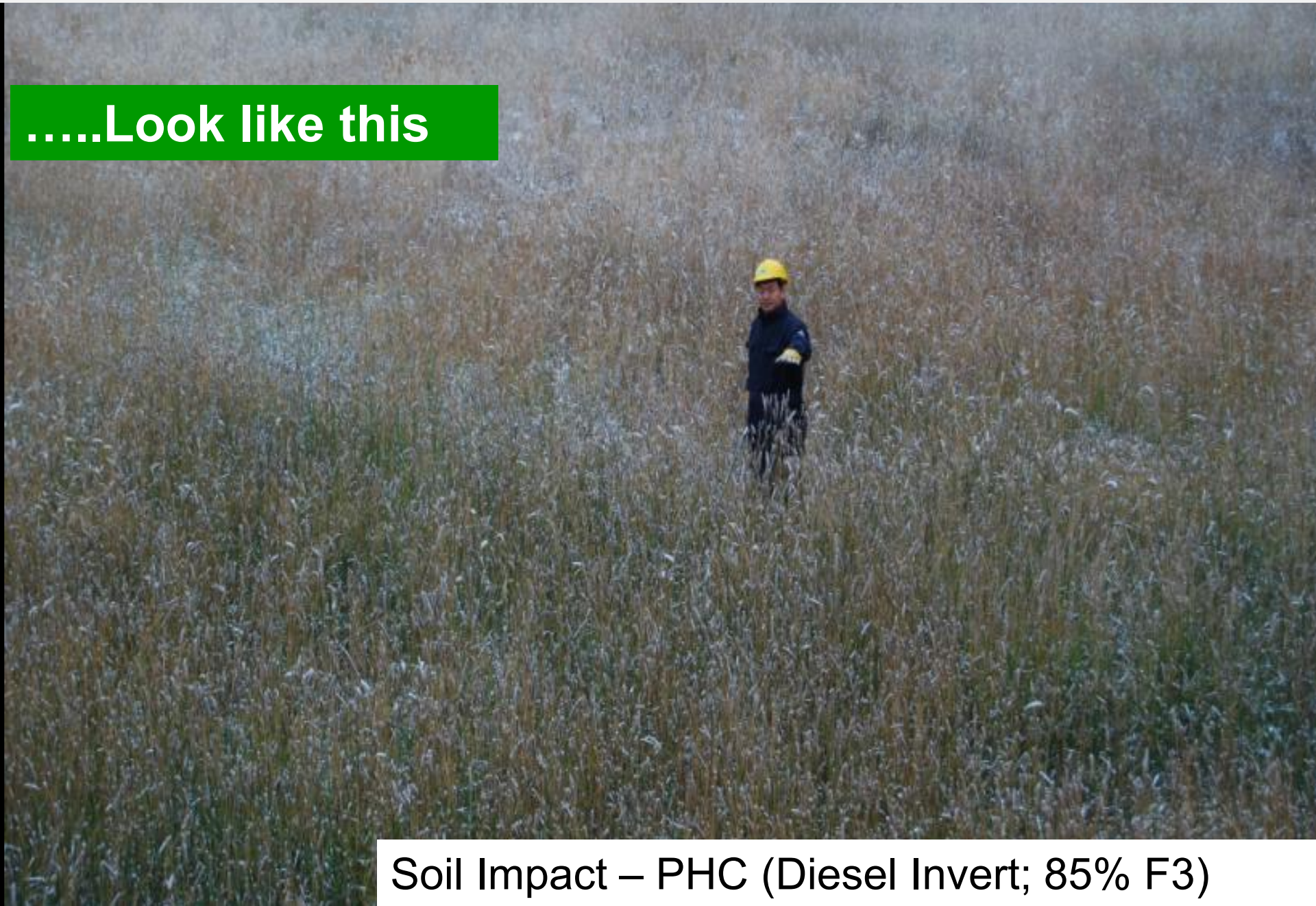
Edson, AB – Before site prep and seeding

PEPS Deployment at Edson, AB

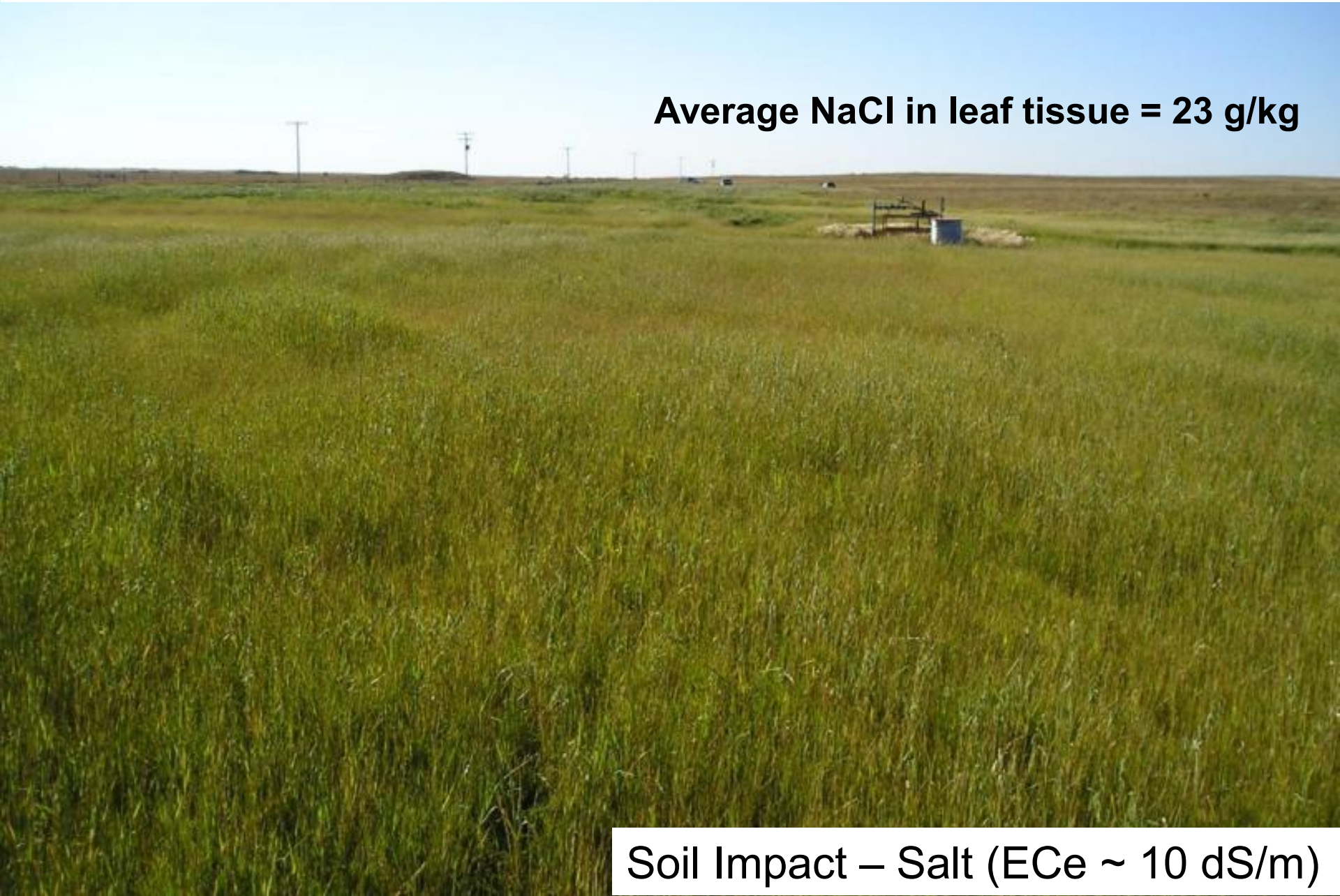


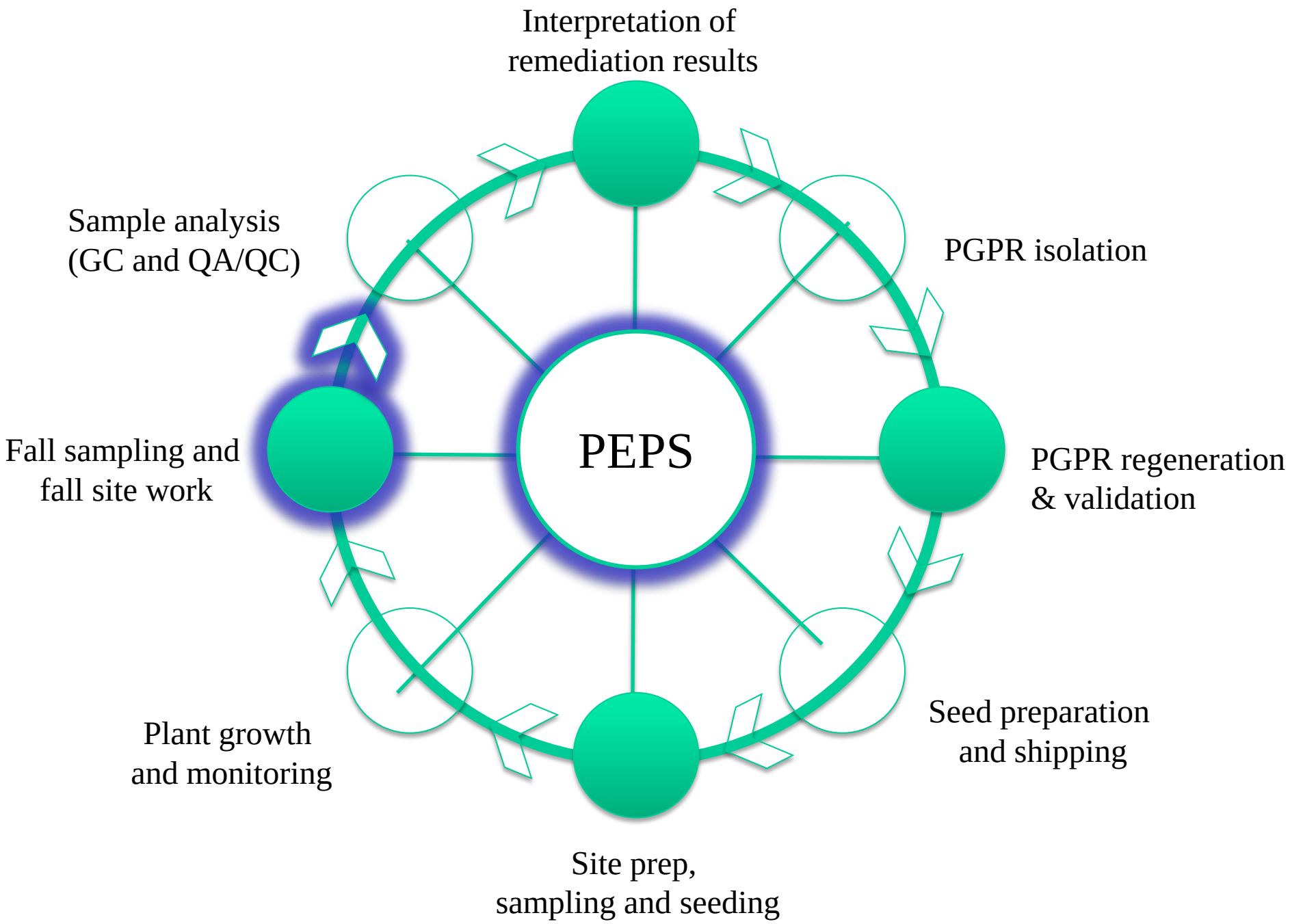
Weyburn, SK - 2: Before site prep and seeding deployment

Weyburn, SK - 2: PEPS deployment – One month

Soil Impact – Salt (ECe ~ 10 dS/m)

Weyburn, SK - 2: PEPS deployment – 3 Months

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Swathing/Mowing

Bailing

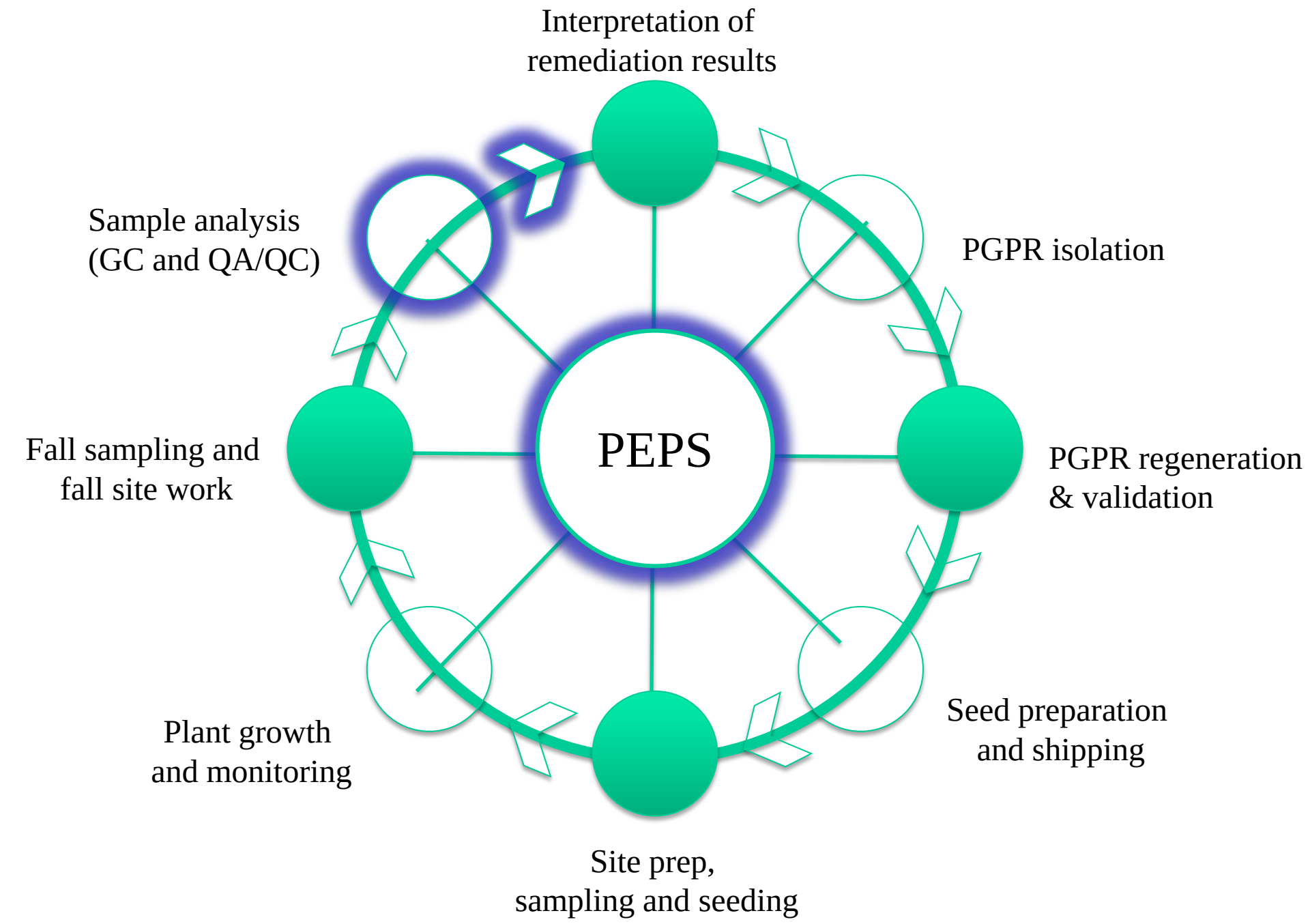
At salt sites, cut grass is removed from the site

- ~ 4000 lbs. of grass were removed from this site
- At remote sites - removal by helicopter
- At PHC sites, grass does not need to be removed

Fall sampling and fall site work

Sampling to get end of season samples

At same sampling points as used at beginning of season.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Sample Analysis

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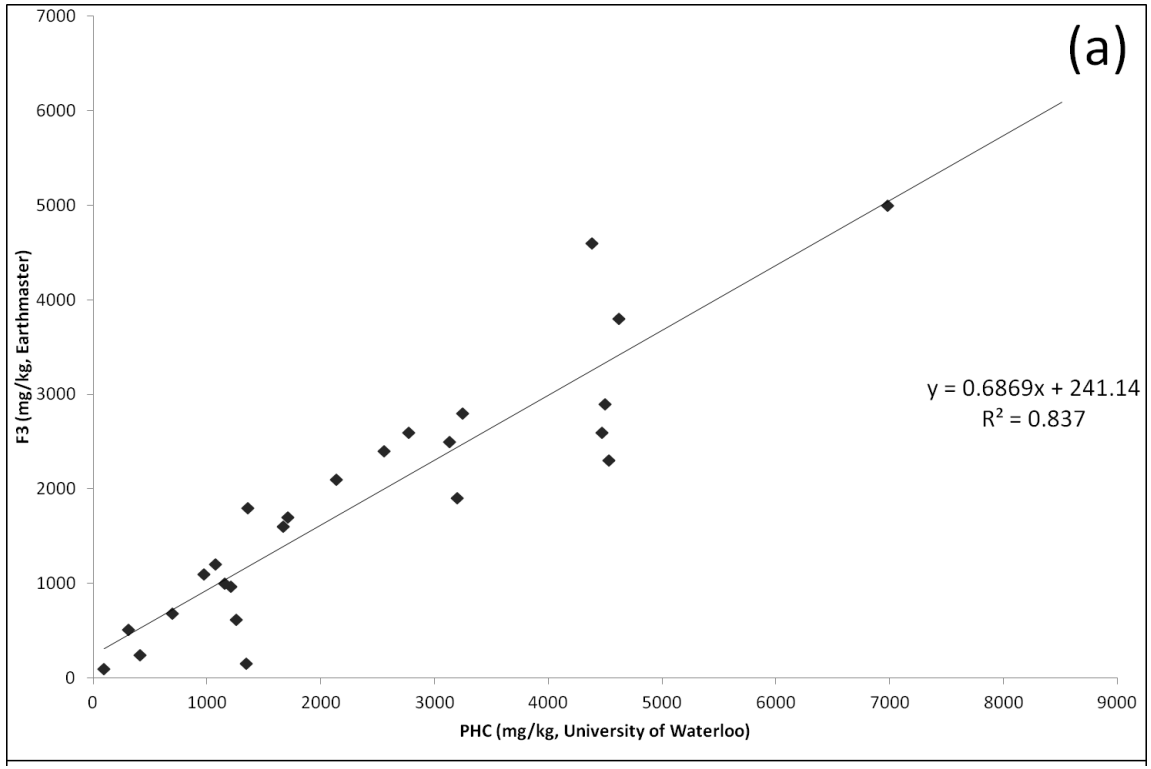
- Analysis of PHC and salt impacted soils
- Soil PHC – CCME GC method
- Soil Salt – ECe, SAR, Na and Cl
- Tissue Salt: Analysis of plant samples to assess plant uptake of salt

QA/QC Analysis PHC

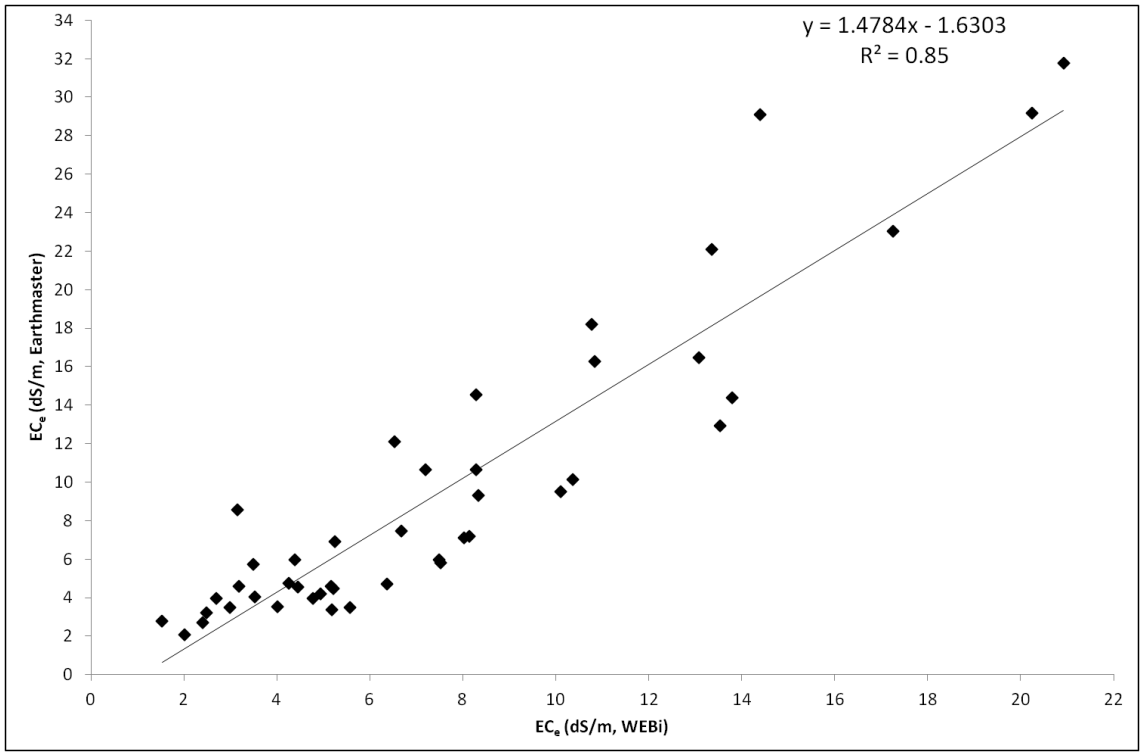


PHC samples are analyzed in at least two laboratories

Data sets compared to assure data quality

Only if correlations are strong are the data accepted

QA/QC Analysis Salt



Salt samples are analyzed in at least two laboratories

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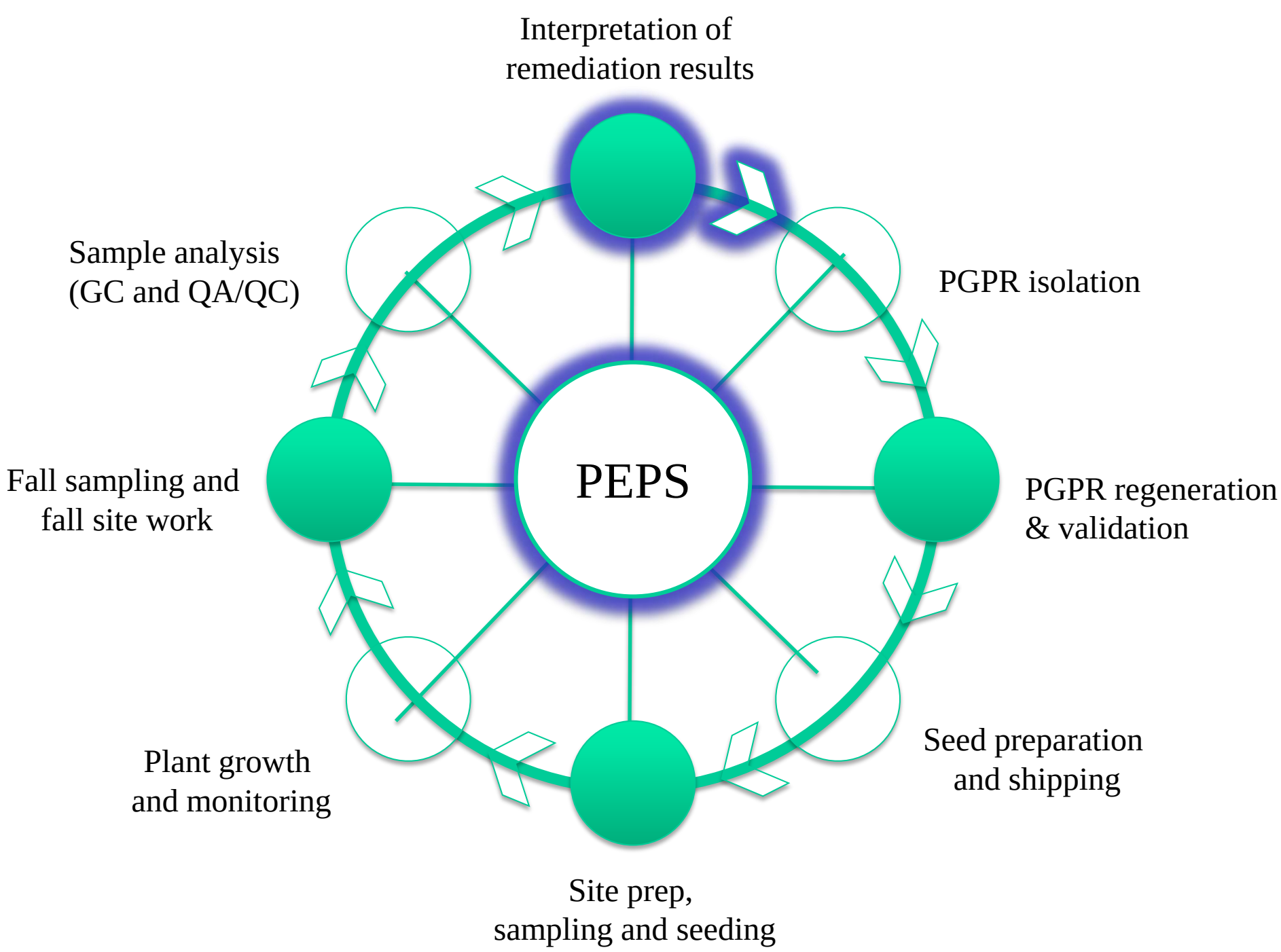
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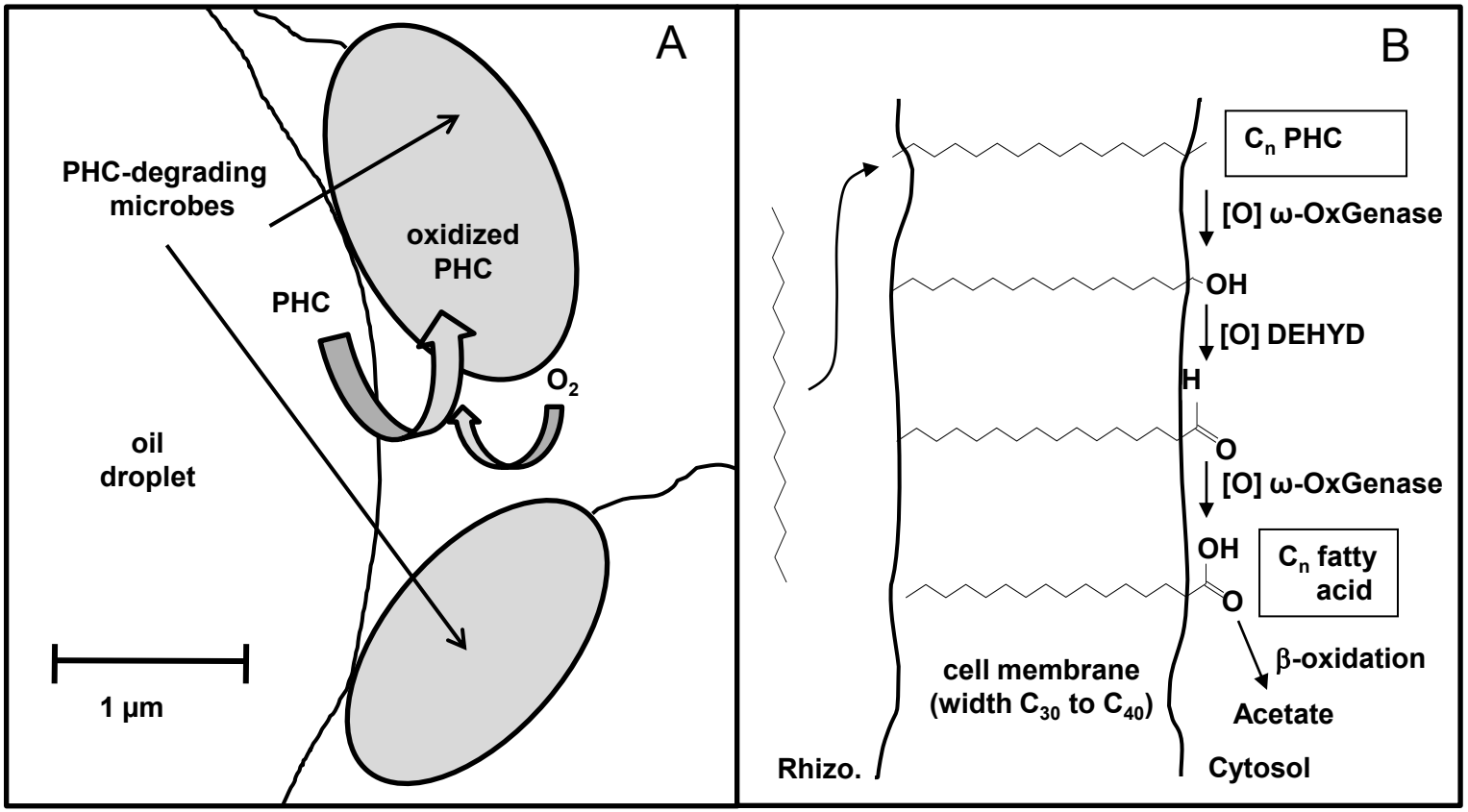
Phytoremediation of PHC

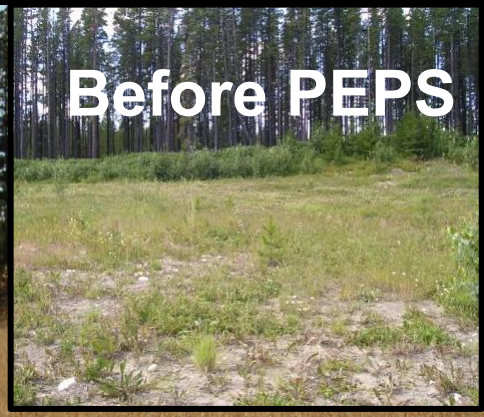
(A) Microbial aerobic PHC degradation – rhizosphere supported by plants

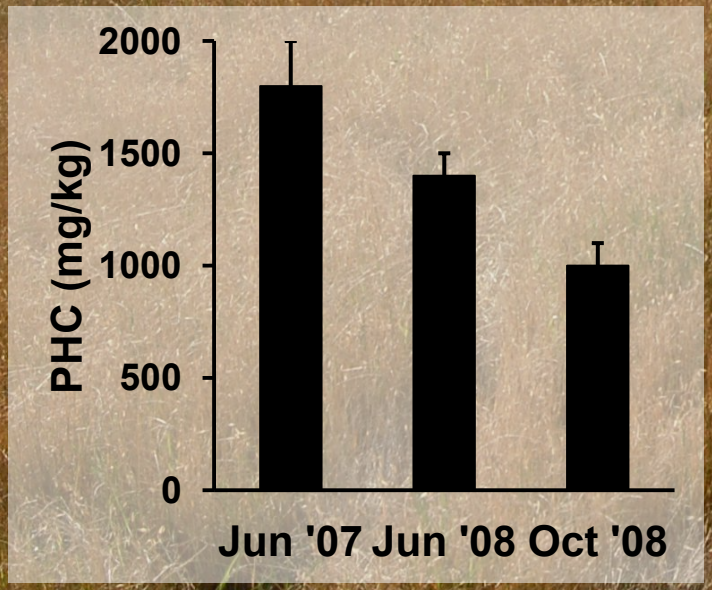
(B) Possible microbial oxygenation pathway of PHC to form a fatty acid



PEPS Deployment at Edson, AB

After PEPS





Interpretation of remediation data

Example of a Completed Site

All 10 sampling points below criteria after remediation

Full Scale PEPS Deployment at Typical PHC Sites

Site	Analysis	Date	Average (mg/kg)	% Remediation	Notes
Completed Sites					
Edson	CCME F3	Spring 2007	1500	33.33%	5 of 10 sample points above Tier 1 criteria
	CCME F3	Fall 2008	1000		All sample points met Tier 1 criteria
Hinton 2	CCME F3	Spring 2007	900	44.44%	6 of 15 sample points above criteria
	CCME F3	Fall 2008	500		All sample points met Tier 1 criteria
Dawson 1	EPH(C10-19)	Spring 2009	6500	91.54%	12 of 12 sample points above Tier 1 criteria
	EPH(C10-19)	Fall 2011	550		1 of 12 sample points above Tier 1 criteria
	EPH(C19-32)	Spring 2009	2500	72.00%	11 of 12 sample points above Tier 1 criteria
	EPH(C19-32)	Fall 2011	700		All sample points met Tier 1 criteria
Peace River	CCME F3	Spring 2007	900	78.89%	4 of 11 sample points above Tier 1 criteria
	CCME F3	Fall 2008	190		All sample points met Tier 1 criteria
Quebec City	CCME F3	Spring 2009	550	49.09%	3 of 3 sample points above criteria
	CCME F3	Fall 2009	280		All sample points met Tier 1 criteria
Sites in Progress					
Hinton 1	CCME F2	Spring 2010	1100	77.27%	10 of 10 sample points above Tier 1 criteria
	CCME F2	Fall 2010	250		6 of 10 sample points above Tier 1 criteria
	CCME F3	Spring 2010	3200	56.25%	9 of 10 sample points above Tier 1 criteria
	CCME F3	Fall 2010	1400		3 of 10 sample points above Tier 1 criteria
Swan Hills	CCME F2	Spring 2009	1400	78.57%	8 of 8 sample points above Tier 1 criteria
	CCME F2	Fall 2010	300		4 of 8 sample points above Tier 1 criteria
	CCME F3	Spring 2009	2550	64.71%	7 of 8 sample points above Tier 1 criteria
	CCME F3	Fall 2010	900		1 of 8 sample points above Tier 1 criteria
Dawson 2	EPH(C10-19)	Spring 2009	6500	46.15%	15 of 15 sample points above Tier 1 criteria
	EPH(C10-19)	Fall 2011	3500		8 of 15 sample points above Tier 1 criteria
	EPH(C19-32)	Spring 2009	700	42.86%	3 of 15 sample points above Tier 1 criteria
	EPH(C19-32)	Fall 2011	400		All sample point met Tier 1 criteria
Dawson 3	EPH(C10-19)	Spring 2009	7000	81.43%	11 of 12 sample points above Tier 1 criteria
	EPH(C10-19)	Fall 2011	1300		5 of 15 sample points above Tier 1 criteria
	EPH(C19-32)	Spring 2009	3500	57.14%	12 of 12 sample points above Tier 1 criteria
	EPH(C19-32)	Fall 2011	1500		6 of 12 sample points above Tier 1 criteria
Beaver River	EPH(C10-19)	Spring 2010	1600	25.00%	8 of 20 sample points above Tier 1 criteria
	EPH(C10-19)	Fall 2010	1200		6 of 20 sample points above Tier 1 criteria
	EPH(C19-32)	Spring 2010	850	35.29%	8 of 20 sample points above Tier 1 criteria
	EPH(C19-32)	Fall 2010	550		3 of 20 sample points above Tier 1 criteria

Examples of Completed Sites

Examples of Sites in progress

Average Remediation = 34 % per year

For salt: NaCl in leaves – leaves removed from the site

2 to 4 ha site - 500 kg of salt (NaCl) off the site in the plants per year

Full Scale PEPS Deployment at Typical Salt Sites

Site	Analysis	Date	Average (dS/m)	% Remediation
Completed Sites				
Nota	ECe	Spring 2008	7.7	70.13%
	ECe	Fall 2010	2.3	
Provost	ECe	Spring 2009	14.5	44.83%
	ECe	Fall 2009	8	
Sites in Progress				
Weyburn 1	ECe	Fall 2010	13.5	22.22%
	ECe	Fall 2011	10.5	
Weyburn 2	ECe	Fall 2010	6.9	14.49%
	ECe	Fall 2011	5.9	
Weyburn 3	ECe	Fall 2010	13.5	10.37%
	ECe	Fall 2011	12.1	
Weyburn 4	ECe	Fall 2010	14.3	11.89%
	ECe	Fall 2011	12.6	
Red Earth	ECe	North, Sp 2010	5.2	13.46%
	ECe	North, F 2011	4.5	
	ECe	South, Sp 2010	4.2	9.52%
	ECe	South, F 2011	3.8	
Kindersley	ECe	Spring 2008	5.5	27.27%
	ECe	Fall 2009	4	
Cannigton Manor	ECe	Spring 2007	17.6	32.95%
	ECe	Fall 2008	11.8	

Examples of Completed Sites

Examples of Sites in progress

Why Use PEPS?

- Scientifically proven and tested SOP
- Verified methods for PHC and/or salt impacted sites.
- Remediations at all sites have been successful; > 30 sites.
- Phytoremediation costs < half the cost of landfilling.
- Liability is reduced, not transferred to a landfill.
- Cost effective at remote sites.
- Enhanced CCME BOC method – phytoremediation will meet Tier 1 criteria.
- Tier 2 approach – will work – After PEPS brings F3 levels ≤ 2500 mg/kg no plant toxicity.

Thank you

Please visit us at the Earthmaster booth for more information

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