FAME Profiles of *Rhizobium leguminosarum* Strains Isolated From Saskatchewan Soils. K.E. DUNFIELD*, L.J.C. XAVIER, and J.J. GERMIDA, University of Saskatchewan, Saskatoon, Sask., Canada.

Introduction. Inoculating lentil (Lens *esculentu* L.) and pea (Pisum *sativum* L.) with commercial *Rhizobium Zeguminosarum* inoculants is a common practice in Saskatchewan. However, little is known about populations of native rhizobia, or how these populations affect inoculant performance. Identification of rhizobia generally involves their isolation on selective media from surface sterilized nodules (Vincent, 1970), and further characterization using conventional methods such as nodulation assay, utilization of carbon substrates, bacteriophage typing, and DNA analysis (Hemandez-Lucas *et al.*, 1995). Recently, Jarvis and Tighe (1994) analyzed the cellular fatty acid methyl esters (FAME) of 123 *Rhizobium* strains including 6 unknowns and concluded that *Rhizobium* species can be accurately identified based on their FAMEs.

Objectives. This study generated a *Rhizobium leguminosarum* FAME profile library based on a collection of known reference strains. The library was used as a diagnostic tool to identify putative soil rhizobia isolated from field grown lentil and pea, and assessed as a way of identifying nodule occupants.

Results and Discussion. The FAME profile library for *R. Zeguminosarum* biovars *viceae, phaseoli, and trifolii was* generated using 10 reference strains, including, three ATCC strains, three commercial inoculant strains and four known strains from our lab collection. Forty-four putative rhizobia were isolated from nodules of field-grown lentil and pea and were used for comparison to the library.

The FAME profile library was a useful tool to study putative rhizobia isolated from field grown plants, as 33 of 42 isolates were identified to be *R. Zeguminosarum.* Eleven of 12 putative strains that nodulated pea in a Leonard jar were identified as *R. Zeguminosarum* bv. *viceae* by the FAME library (Table 1). Strains identified as other rhizobia by the library did not nodulate pea.

Table. 1 Identification, nodulation ability and ARA of several putative rhizobia strains isolated from field grown pea.

Culture ID	Nodulation	ARA[a]	Similarity Index	Strain Identification
LX01	+	3	0.72	*R. leguminosarum* bv. *viceae*
LX05	+	3	0.86	*R. leguminosarum* bv. *viceae*
LX06	+	3	0.80	*R. leguminosarum* bv. *viceae*
LX07	+	5	0.86	*R. leguminosarum* bv. *viceae*
LX08	+	4	0.74	*R. leguminosarum* bv. *viceae*
LX09	+	4	0.56	*R. leguminosarum* bv. *viceae*
LX11	+	2	0.30	*R. leguminosarum* bv. *viceae*
LX17	+	5	0.61	*R. leguminosarum* bv. *viceae*
LX18	+	5	0.87	*R. leguminosarum* bv. *viceae*
LX19	+	4	0.58	*R. leguminosarum* bv. *viceae*
LX20	+	5	0.56	*R. leguminosarum* bv. *viceae*
LX02		0	0.79	*R. leguminosarum* bv. *trifolii*
LX10		0	0.35	*R. meliloti*
LX12		0	0.02	*X. maltophilia*
LX13	+/- (?)	1		No match

[a]Relative acetylene reduction activity (ARA) with 5 as the highest level and 0 as not detected.

Generation of nodule FAME profiles might be used to establish the identity of nodule occupants. Preliminary experiments conducted with pea inoculated with pure cultures of *R. leguminosarum* strains and grown in Leonard jars revealed that fatty acid profiles could be obtained from uninoculated (control) pea roots and nodules. A FAME profile of the nodule occupant was deduced by subtracting the percentage of control fatty acids from the nodule fatty acids. Fatty acids present in the nodule occupant and the inoculant, but not the control may be indicative of the inoculant strain (Figure 1).

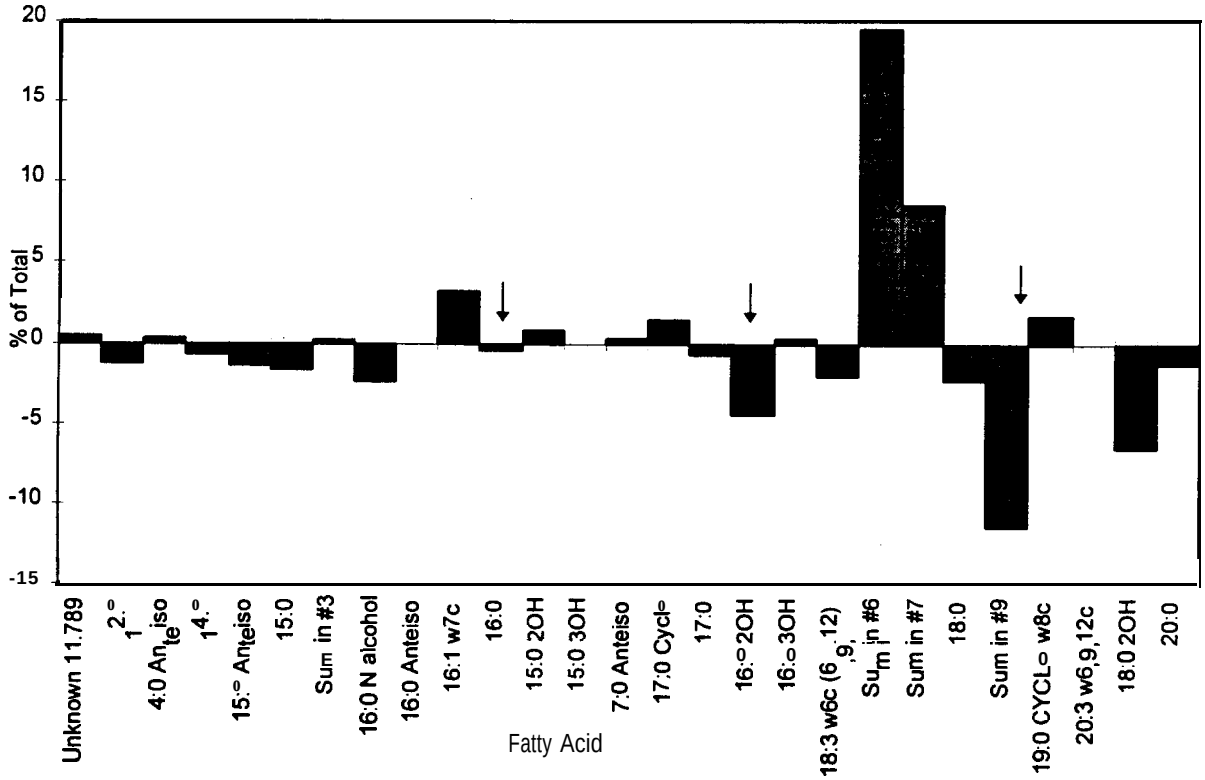

Figure 1. Nodule occupant profile of *R. leguminosarum* bv. *viceae strain C* 1. Arrows indicate feature fatty acids found only in the occupant and inoculant FAME profile.

In summary, a *Rhizobium* FAME profile library was developed and successfully used to distinguish between rhizobia and other soil bacteria. In addition, our results show that feature fatty acids (i.e., strain or isolate-specific) found in the nodule and inoculant FAME profiles might be used for the identification of nodule occupants. The *Rhizobium* FAME library is not complete, and more entries will be added to enhance it's utility for ecological studies.

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