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## The Influence of Potato Cultivar on Lipid Content and Fecundity of Bolivian and British Populations of *Globodera rostochiensis*

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**Abstract:** The influence of host cultivar on the lipid levels provided by a female to her progeny was investigated with Oil Red O stain and a quantitative image analyzer. A population of *Globodera rostochiensis* was multiplied at Toralapa Field Station in Bolivia on 25 different potato cultivars grown in that country. The mean neutral lipid content of newly formed second-stage juveniles varied significantly with cultivar over a 200% range. The corresponding range was only 18% and 28% for the same Bolivian and a UK population of *G. rostochiensis*, respectively, when both completed reproduction concurrently on 10 pot-grown European cultivars in the United Kingdom. Egg numbers per female varied with host for Bolivian cultivars that lack known partial resistance to *Globodera* spp. There was a 15-fold range between the most and least fecund nematode-host combinations (Kosi and Gendarme). The Bolivian *G. rostochiensis* population showed only a 2-fold range in mean eggs per cyst when grown on European cultivars in the UK. The fatty acid profiles of lipids from Bolivian *G. rostochiensis* cysts reared on Bolivian potato cultivars were dominated by C<sub>20</sub> (37–64%) and C<sub>18</sub> (28–46%) fatty acids and ranged from C<sub>14</sub> to C<sub>22</sub>. The three major fatty acids detected were C<sub>20:4</sub>, C<sub>20:1</sub>, and C<sub>18:1</sub>. Few differences between cultivars were observed. For a UK population of *G. rostochiensis* reared on ssp. *tuberosum*, higher relative percentages of C<sub>18</sub> and monounsaturated fatty acids and lower relative percentages of C<sub>20</sub> and polyunsaturated fatty acids were found.

**Key words:** Bolivia, fatty acid, fecundity, *Globodera rostochiensis*, lipids, nematode, neutral lipid, Oil Red O, potato, potato cyst nematode.

Lipids and carbohydrates are the main energy reserves for plant-parasitic nematodes (Barrett, 1981). The extent of neutral lipid reserves influences the duration of pre-hatch survival and post-hatch infectivity of potato cyst nematode (PCN) second-stage juveniles (J2) (Robinson et al., 1985; Storey, 1984). Neutral lipids from unhatched and freshly hatched J2 of PCN are more than 70–80% of total lipid (Holz et al., 1997, 1998a, 1998b). Their quantity within the J2

is correlated with both infectivity and motility in J2 of *G. rostochiensis* (Robinson et al., 1985). Unhatched, dormant *G. pallida* J2 use their lipid reserves in UK field soils about 80 times slower than hatched J2 stored in potato root diffusate at 20 °C. They still have 50% of their initial lipid reserves after 7.5 years in the field (Storey, 1984).

The importance of lipid reserves to J2 of PCN led us to investigate the influence of host cultivar on the extent of neutral lipid levels provided by females of Bolivian and British populations to progeny. The potato cyst nematodes, *Globodera rostochiensis* and *G. pallida*, and false root-knot nematode, *Nacobbus aberrans*, are the top-ranked biotic stresses for potato in the Bolivian Andes (Programa de Investigación de la Papa, 1997). This work is based on Oil Red O staining and quantitative image analysis. We also determined the fatty acid composition of the main lipid classes in Bolivian *G. rostochiensis* cysts grown on Bolivian potato culti-

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vars. In addition, egg numbers per female were evaluated for a range of different cultivars.

#### MATERIALS AND METHODS

**Cysts:** In the first experiment, all PCN cysts came from single-generation cultures grown in pots under glasshouse conditions. The plants were grown at Toralapa Field Station (latitude 18 °S), Department of Cochabamba, Bolivia, at an altitude of 3,300 m. A total of 25 potato (*Solanum tuberosum* L.) cultivars available to growers in Bolivia (Table 1) were planted in soil containing 30 to 50 eggs of *G. rostochiensis*/gram, a common preplant density in Bolivia (Programa de Investigación de la Papa, 1997). Cysts were collected after potato harvest, air-dried, and sent airmail in plastic vials to Leeds, United Kingdom, where they were stored at 4°C until use. About 400 mg dry weight each of these cysts reared on cultivars Waycha, Puca Toralapa, and Runa Toralapa were used for the analysis of fatty acid profiles by gas chromatography (see below). Tests using both PCR-techniques and stilet measurements indicated that the Bolivian PCN cysts used were more than 90% *G. rostochiensis*. Because *G. pallida* was not positively identified in the samples, it likely was not present at a density that could influence the results.

Cysts from two different sources were used in a second experiment. The Bolivian *G. rostochiensis* population used previously was multiplied as above on Waycha, a subspecies *andigena* cultivar, and cysts were sent to the United Kingdom. The second population of *G. rostochiensis* was from the United Kingdom and was cultured on cv. Désirée (ssp. *tuberosum*) in glasshouse pot cultures in Leeds, United Kingdom. Ten potato cultivars were grown in 20-cm-diam. pots with non-sterile soil in a plastic tunnel outdoors. The cultivars were Duke of York (First Early [FE]), Foremost (FE), Maris Barr (FE), Wilja (Second Early [SE]), Catriona (SE), Maris Peer (SE), Désirée (Main Crop (MC)), Majestic (MC), King Edward (MC), and Pentland Crown (MC). The soil in 3

pots of each cultivar was infested with 40 cysts/pot of the British population and another 3 pots were infested with the same cyst number of the Bolivian population to provide ca. 5 eggs/gram soil for both populations. Cysts were extracted 110 days after infestation with a Fenwick can (Shepherd, 1970), air-dried, and stored at 4 °C until use.

**Neutral lipid content:** Oil Red O was used to stain the neutral lipids of *G. rostochiensis* J2 (Croll, 1972). Ten cysts per treatment were soaked for 4 days, then crushed gently between two glass slides to release juveniles. Oil Red O solution was warmed in a water bath at 60 °C for 30 minutes. Approximately 2 ml of 60 °C ORO solution was used to wash the crushed cysts and liberated juveniles of each treatment from the glass slides into pre-warmed 10-ml polythene centrifuge tubes. Each population set was crushed and washed separately. The centrifuge tubes were placed in a 60 °C water bath for a 30-minute staining period. The J2 were then centrifuged at 1,500g for 5 minutes at 20 °C. Fluid was reduced to 50 µl, then J2 were resuspended in 1 ml of 70% ethanol:glycerol (1:1 v/v). The centrifugation was repeated three times or until the solution was colorless to the eye. Finally, the supernatant was removed and 0.1 ml of glycerol was used to resuspend the J2. The nematodes were mounted onto a microscope slide in the glycerol under a coverslip sealed with nail varnish.

**Image analysis:** The ORO-stained J2 were analyzed under a Leica microscope fitted with a color camera (Kappa CF 15 MCC), which was connected to a frame grabber. Analysis was controlled with Quantimet 500C software (Leica, London, UK). Each image pixel represented 0.449 mm as calibrated with a micrometer slide. A pair of thresholds was set for each of three color components (red, blue, and green) for color detection. The thresholds were selected by identifying one or several well-stained regions within the intestine of a juvenile, after which at least 60 whole J2 derived from 10 cysts were analyzed per cultivar.

**Lipid analysis:** The methods used to deter-

mine lipid and fatty acid composition were as described in Holz et al. (1997). Whole Bolivian *G. rostochiensis* cysts, reared on cultivars Puca Toralapa, Runa Toralapa, and Waycha in Bolivia, were homogenized with a microhomogenizer on dry ice and freeze-dried for 48 hours. Nematode lipids were extracted with a modified (Bailey, 1970) method of Folch et al. (1957). About 20% of each sample was retained for total lipid analysis and the rest was separated into different lipid classes by solid phase chromatography (Figlewicz et al., 1985; Kaluzny et al., 1985). Aminopropyl-bonded columns (Bond Elut, Varian Analytical Instruments, Valencia, CA) and several solvents of different polarities were used to separate the sample into neutral lipids, free fatty acids, and non-acidic phospholipids (Figlewicz et al., 1985; Kaluzny et al., 1985). Acidic phospholipids would not be eluted by this method (Christie, 1992). The samples were then dried, re-weighed, dissolved in chloroform, and stored at  $-20^{\circ}\text{C}$  in glass vials purged with nitrogen.

Lipids (except the free fatty acid fraction) were hydrolyzed with 0.5 M NaOH in methanol (1 ml) under reflux in a water bath at  $100^{\circ}\text{C}$ . Fatty acid methyl esters (FAMEs) were obtained by first incubating lipids with 1 ml boron trifluoride under reflux for 3 minutes; hexane (1 ml) was then added and the samples were left under reflux for 1 minute (Morrison and Smith, 1964). The FAMEs were analyzed with a Varian Vista 401 gas chromatograph (GC) equipped with a polar Carbowax column (30-m length, 0.32-mm internal diam., 0.25- $\mu\text{m}$  film). Retention times of FAMEs were compared with authentic standards (Restek, Maidenhead, UK; Matreya, Pleasant Gap, PA). Two GC runs were conducted per sample with two samples per treatment.

*Statistical analysis:* The statistical programs SPSS 6.1 and Excel 5 were used. Confidence was tested at the 5% level unless otherwise stated. Duncan's multiple-range test was used to determine homogeneous subsets, in which the differences among means are not significantly different.

## RESULTS

*Bolivian pot experiment:* Cysts from cultivars Karnico and Mondial were discarded because they were too few and too small for this study, presumably because of host-plant resistance to *G. rostochiensis*. The stained area of neutral lipid was measured in 1,380 J2 produced by females on the remaining 23 cultivars (Table 1). The overall mean value was  $1,405 \pm 11 \mu\text{m}^2/\text{J2}$ . The mean neutral lipid area ranged from  $887 \pm 41 \mu\text{m}^2$  (cv. India) to  $1,858 \pm 61 \mu\text{m}^2$  (cv. Waycha), an approximately 2-fold difference (Fig. 1A). Neutral lipid area varied widely among cultivars; cv. India had a significantly lower value ( $P < 0.05$ ) than all other cultivars.

Relative differences in the mean number of eggs per female gave a larger range of values than occurred for neutral lipid con-

TABLE 1. Potato cultivars used in a pot experiment at Toralapa Field Station, Cochabamba, Bolivia.

Cultivar	<i>Solanum tuberosum</i> subspecies	Resistance
Agria	hybrid <sup>a</sup>	PCN <sup>c</sup> (Ro1) <sup>d</sup>
Alpha	ssp. <i>tuberosum</i>	—
Chaposa	hybrid	<i>Phytophthora infestans</i>
Colombiana	hybrid	—
Désirée	ssp. <i>tuberosum</i>	—
Gendarme	ssp. <i>andigena</i>	<i>Nacobbus aberrans</i>
Goyllu	ssp. <i>andigena</i>	—
Imilla Blanca	ssp. <i>andigena</i>	—
Imilla Negra	ssp. <i>andigena</i>	—
India	hybrid	<i>P. infestans</i>
Jaspe	hybrid	<i>Nacobbus</i> ; <i>P. infestans</i>
Kallpa Runa	ssp. <i>andigena</i>	—
Karnico	hybrid	PCN (Ro1,2,3; Pa2)
Kosi	ssp. <i>andigena</i>	—
Lucky	<i>S. juzepczukii</i> <sup>b</sup>	—
Maria Huanca	hybrid	PCN (P4A, P5A)
Mondial	hybrid	PCN (Ro1)
Perla	ssp. <i>tuberosum</i>	<i>P. infestans</i>
Puca Toralapa	hybrid	—
Runa Toralapa	hybrid	<i>P. infestans</i>
Robusta	hybrid	<i>P. infestans</i>
Runa	ssp. <i>andigena</i>	—
Sani	ssp. <i>andigena</i>	—
Waycha	ssp. <i>andigena</i>	—
Yungay	hybrid	—

<sup>a</sup> Hybrid, both ssp. *tuberosum* and ssp. *andigena* parentage.

<sup>b</sup> *S. juzepczukii*, hybrid between *S. acaule* and *S. stenotomum*.

<sup>c</sup> PCN = potato cyst nematodes.

<sup>d</sup> Pathotype.

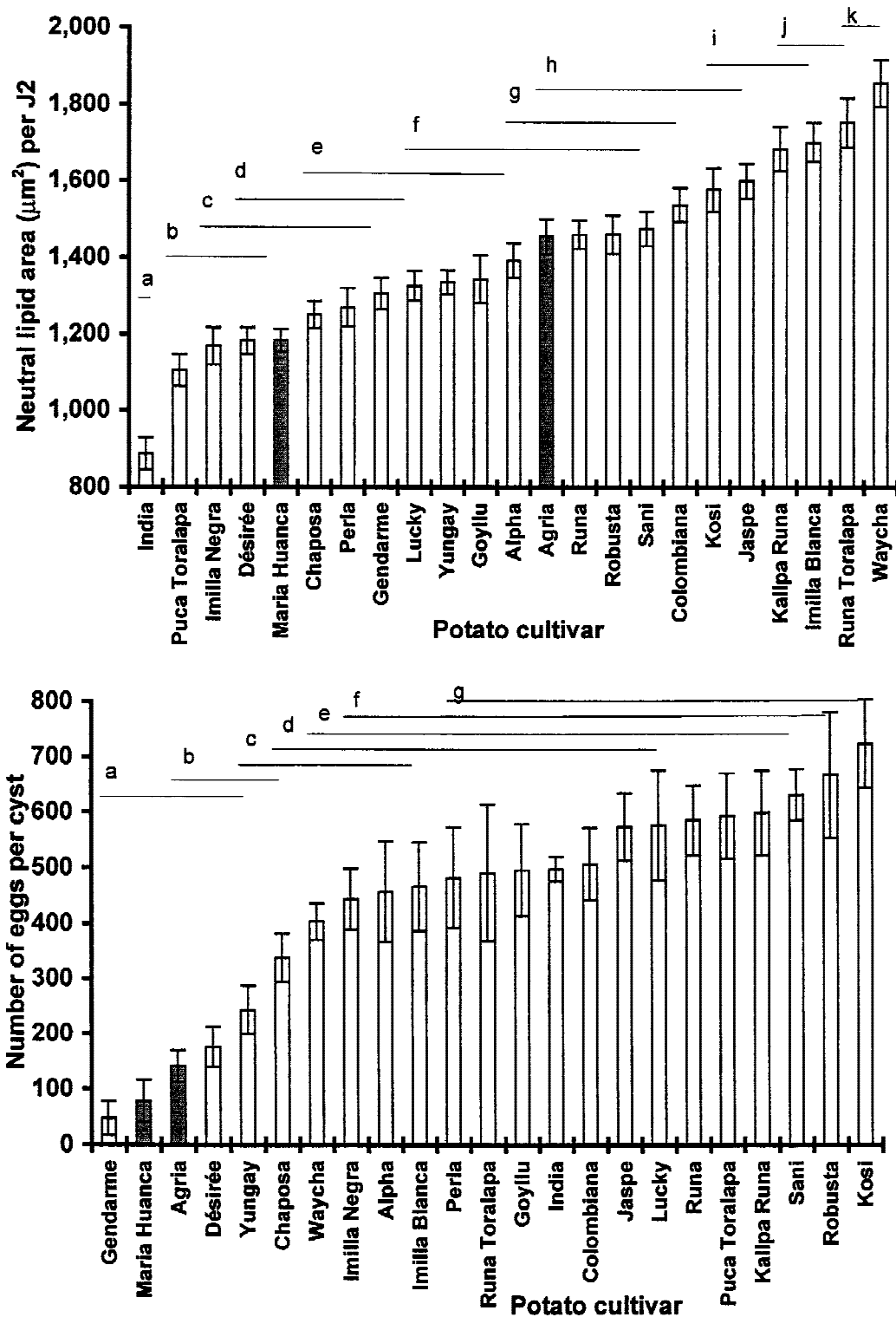


FIG. 1. A) Mean neutral lipid content of second-stage juveniles (J2) of *Globodera rostochiensis* reared on 23 Bolivian potato cultivars in pot experiments at Toralapa Field Station, Cochabamba, Bolivia. B) Mean number of eggs per cyst of *G. rostochiensis* reared on 23 Bolivian potato cultivars in pot experiments at Toralapa Field Station, Cochabamba, Bolivia. Bars represent standard error of means. Gray columns indicate cultivars with known resistance to *G. rostochiensis*. Columns grouped by a common horizontal line do not differ ( $P > 0.05$ ). Cultivars Alpha, Désirée, and Perla are ssp. *tuberosum*; all other cultivars are of Bolivian origin.

tent (Fig. 1B). The mean egg number per cyst was  $439 \pm 22$  eggs, but values showed a 15-fold range among cultivars from  $47 \pm 30$  eggs for Gendarme to  $724 \pm 94$  eggs for Kosi. Cysts from most cultivars contained more than 400 eggs, with less than 200 eggs/cyst found only for Gendarme, Maria Huanca, Agria, and Désirée.

*British pot experiment:* The mean neutral lipid area per J2 for more than 650 J2 from Bolivian cysts, grown in the United Kingdom on ssp. *tuberosum* cultivars, was  $1,587 \pm 15 \mu\text{m}^2$  (Fig. 2A). The means for different cultivars ranged from  $1,467 \pm 38 \mu\text{m}^2$  for Majestic to  $1,731 \pm 46 \mu\text{m}^2$  for Foremost. These values represent a range difference of 18% with three homogeneous subsets (Fig. 2A). Means obtained from UK cysts grown on the same cultivars provided a 28% range of values, from  $1,286 \pm 44 \mu\text{m}^2$  for Maris Peer to  $1,640 \pm 40 \mu\text{m}^2$  for Duke of York (Fig. 2B). The overall mean was  $1,492 \pm 13 \mu\text{m}^2$  neutral lipid area/J2 and four homogeneous subsets were found (Fig. 2B). In eight of the ten cultivars tested, mean neutral lipid area was greater in the Bolivian rather than British populations (Figs. 2A,B).

The number of eggs per cyst was counted for 10 Bolivian cysts per cultivar. The mean was  $531 \pm 15$  eggs/cyst, ranging from  $292 \pm 28$  eggs (cv. Catriona) to  $627 \pm 30$  eggs (cv. Pentland Crown) — a 2.2-fold difference (Fig. 3). Results for Catriona differed significantly from all other cultivars. Three homogeneous subsets were identified ( $P < 0.05$ ; Fig. 3).

*Lipid analysis:* The majority of total lipid was neutral lipid (>80%), with much less phospholipid and only a small fraction of free fatty acids. Altogether, 17 fatty acids could be identified, ranging from  $\text{C}_{14}$  to  $\text{C}_{22}$ . The fatty acid profiles were dominated by  $\text{C}_{20}$  (37–64%) and  $\text{C}_{18}$  (28–46%) (Table 2). About 40% each of the fatty acids in the total lipids were mono- and polyunsaturated with the remaining 20% being saturated. The three major fatty acids,  $\text{C}_{20:4}$ ,  $\text{C}_{20:1}$ , and  $\text{C}_{18:1}$ , accounted for ca 50–70% (Table 2).

The fatty acid profiles of the total lipid fractions of *G. rostochiensis* cysts reared on potato cultivars Puca Toralapa, Runa To-

ralapa, and Waycha were compared. Few differences were observed among cultivars (Table 2). The same degree of similarity was found within the other lipid classes. Exceptions were  $\text{C}_{18:1}$  in total and neutral lipids and  $\text{C}_{20}$  in free fatty acids, which were significantly different (Table 2).

## DISCUSSION

Large and statistically significant differences were found in both the neutral lipid content of J2 and the number of eggs per cyst when the Bolivian population of *G. rostochiensis* was reared on different potato cultivars in Bolivia. Nematodes reared on the three ssp. *tuberosum* cultivars (Désirée, Perla, and Alpha), grown under short-day conditions at Toralapa, showed a neutral lipid content below the grand mean. However, cysts formed on both Perla and Alpha contained an egg number that was close to the overall mean. Ellenby (1958) found that a shortened day length significantly reduced the number of eggs per cyst in two of three ssp. *tuberosum* cultivars tested. Cultivars of ssp. *tuberosum* appeared only in the five homogeneous groups with the lowest neutral lipid content. The four groups with high neutral lipid content were dominated by hybrids and ssp. *andigena* cultivars.

Cysts of Bolivian isolates reared in UK pot experiments on ssp. *tuberosum* contained significantly more neutral lipid and eggs than did the same isolates reared in pots on different cultivars in Bolivia. This difference was not due to the use of resistant cultivars (Maria Huanca and Agria) in the Bolivian experiment, which caused only a slight decrease in the overall means of neutral lipid content and number of eggs. Presumably, the growing conditions in Leeds favored a larger neutral lipid content increase and greater fecundity in *G. rostochiensis*. Different Pi's used in the Bolivian (30–50 eggs/gram soil) and the Leeds (4–5 eggs/gram) experiments might have affected neutral lipid content and fecundity. However, previous pot experiments with *G. rostochiensis* demonstrated that the density and fecundity of females did not influence the neutral lipid

TABLE 2. Fatty acid composition (%) of lipids from cysts of *Globodera rostochiensis* reared on different potato cultivars.<sup>a</sup>

Fatty acid	Total lipid						Neutral lipid						Free fatty acid						Phospholipid						
	Bolivia			UK			Bolivia			UK			Bolivia			UK			Bolivia			UK			
	Way	PTo	RTTo	PI <sup>c</sup>	Des	For	P2	Way	PTo	PI	Des	For	P2	Way	PTo	PI	Des	For	P2	Way	PTo	PI	Des	For	P2
C <sub>14:0</sub>	0.9	1.0	0.9	0.8	0.6	0.6	0.7	0.7	0.7	0.7	0.7	0.7	0.7	1.6	2.1	2.1	0.9	0.9	0.7	0.6	0.4	0.4	0.8	0.5	0.5
iso-C <sub>15:0</sub>	1.0	0.9	1.0	1.3	1.0	1.0	0.8	1.0	0.8	1.0	1.0	1.0	0.7	1.0	0.5	**	1.6	1.0	1.0	0.7	0.4	0.4	0.6	1.3	1.3
C <sub>16:0</sub>	3.0	4.3	3.3	3.7	3.2	3.3	3.1	3.0	3.1	3.0	3.0	3.0	3.0	8.4	9.9	*	5.0	4.8	*	2.8	4.2	**	3.0	3.2	3.2
C <sub>16:1</sub>	0.4	0.3	0.3	1.4	1.5	***	1.1	1.1	1.1	1.1	1.1	1.1	*	2.6	3.4	*	1.2	1.4	*	0.5	—	**	1.0	1.2	1.2
iso-C <sub>17:0</sub>	0.7	0.6	0.6	0.8	0.7	*	0.6	0.7	0.6	0.7	0.6	0.7	0.7	0.9	0.8	*	1.1	1.0	1.0	0.5	0.4	0.4	0.8	0.7	0.7
C <sub>16:3</sub>	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	1.7	1.2	—	—	—	*
C <sub>18:0</sub>	6.7	6.9	6.6	8.9	8.0	*	6.3	6.5	8.1	6.9	8.1	6.9	8.1	12.3	13.6	—	10.7	10.5	—	6.1	6.5	6.1	7.6	6.4	6.4
C <sub>18:1</sub>	18.8	15.8	16.8	21.2	19.4	*	16.0	15.4	18.2	18.3	18.3	18.3	*	19.5	19.7	—	30.4	26.5	*	25.2	26.3	25.2	28.5	29.3	*
C <sub>18:2</sub>	4.0	3.8	3.7	3.6	3.6	***	3.1	3.3	3.4	3.1	3.1	3.1	3.1	3.6	3.6	—	1.9	1.7	**	10.9	11.3	10.9	6.4	7.4	*
C <sub>18:3</sub>	2.1	2.1	2.1	3.0	3.2	***	2.3	2.2	2.9	2.5	2.5	2.5	2.5	1.1	1.0	—	0.9	0.5	—	1.8	1.9	1.8	2.1	2.8	*
C <sub>20:0</sub>	4.9	4.3	4.4	4.6	5.0	—	3.7	4.1	4.5	4.1	4.5	4.1	*	5.9	5.2	*	4.7	5.7	—	6.6	5.4	6.6	5.9	4.8	4.8
C <sub>20:1</sub>	20.5	22.1	22.4	21.3	20.9	—	21.8	21.8	19.9	20.4	20.4	20.4	*	21.3	21.7	—	36.8	38.2	**	21.1	20.5	21.1	26.1	23.7	23.7
C <sub>20:2</sub>	1.5	1.6	1.0	0.9	1.0	—	1.1	1.1	1.1	0.9	1.1	0.9	0.9	—	—	—	—	0.7	—	0.9	0.9	0.9	0.8	0.6	0.6
C <sub>20:3</sub>	4.6	5.0	4.9	5.0	5.4	—	5.0	4.9	6.2	6.0	6.0	**	**	1.4	1.0	—	—	0.6	—	4.4	4.8	4.4	3.7	4.1	4.1
C <sub>20:4</sub>	28.6	29.1	29.7	22.0	24.7	*	33.0	32.3	27.9	29.5	29.5	*	*	10.6	8.6	—	1.6	2.3	*	11.8	11.9	11.8	10.7	12.4	12.4
C <sub>22:0</sub>	1.4	1.8	1.9	1.4	1.5	**	1.1	1.2	1.4	1.2	1.4	1.2	1.2	9.1	7.8	—	2.4	3.3	*	3.4	3.4	3.4	1.6	1.5	***
C <sub>22:1</sub>	0.9	0.4	0.5	—	0.3	—	0.5	1.0	0.2	0.3	0.3	0.3	0.3	0.8	1.2	—	0.7	0.9	—	0.9	0.6	0.9	0.4	—	—
C <sub>14</sub>	0.9	1.0	0.9	0.8	0.6	—	1.0	0.9	0.7	0.7	0.7	0.7	*	1.6	2.1	—	0.9	0.7	—	0.6	0.4	0.6	0.8	0.5	0.5
C <sub>16</sub>	4.4	5.6	4.6	6.5	5.6	*	4.5	4.6	5.0	5.1	5.1	5.1	*	12.0	13.8	*	7.8	7.3	*	5.8	5.8	5.8	4.6	5.7	5.7
C <sub>18</sub>	32.3	29.2	29.8	37.5	35.0	*	28.4	28.0	33.2	31.6	31.6	*	*	37.3	38.6	*	45.0	40.2	*	44.5	46.4	44.5	45.4	46.6	46.6
C <sub>20</sub>	60.1	62.1	62.4	53.9	57.0	*	64.5	64.2	59.6	61.0	61.0	*	*	39.2	36.5	*	43.2	47.5	*	44.8	43.4	44.8	47.2	45.6	45.6
C <sub>22</sub>	2.3	2.2	2.4	1.4	1.8	*	1.6	2.2	1.5	1.5	1.5	1.5	*	9.9	9.0	—	3.1	4.2	*	4.3	4.0	4.3	2.0	1.5	*
Saturated	18.6	19.9	18.7	21.5	20.0	—	16.8	17.8	19.1	17.7	17.7	17.7	*	39.1	39.8	—	26.5	27.1	**	20.7	20.7	20.7	20.4	18.5	18.5
Monounsaturated	40.6	38.6	39.9	43.9	42.2	*	38.7	38.4	39.4	40.2	40.2	*	*	44.2	46.0	—	69.1	67.0	**	47.7	47.4	47.7	56.0	54.2	*
Polysaturated	40.8	41.6	41.4	34.6	37.8	*	44.5	43.8	41.5	42.1	42.1	*	*	16.7	14.2	—	4.4	5.9	*	31.7	31.9	31.7	23.6	27.3	27.3

<sup>a</sup> Des = cv Désirée (*Solanum tuberosum tuberosum*); UK; For = cv Foremost (*S.t.t.*; UK); Way = cv Waycha (*S.t. andigena*; Bol.); PTo = cv Puca Toralapa (hybrid; Bol.); RTTo = cv Runa Toralapa (hybrid; Bol.).

<sup>b</sup> UK data from Holz et al. (1998c).

<sup>c</sup> PI = Single Factor ANOVA between Bolivian cultivars; P2 = Single Factor ANOVA between Bolivian and UK cultivars.

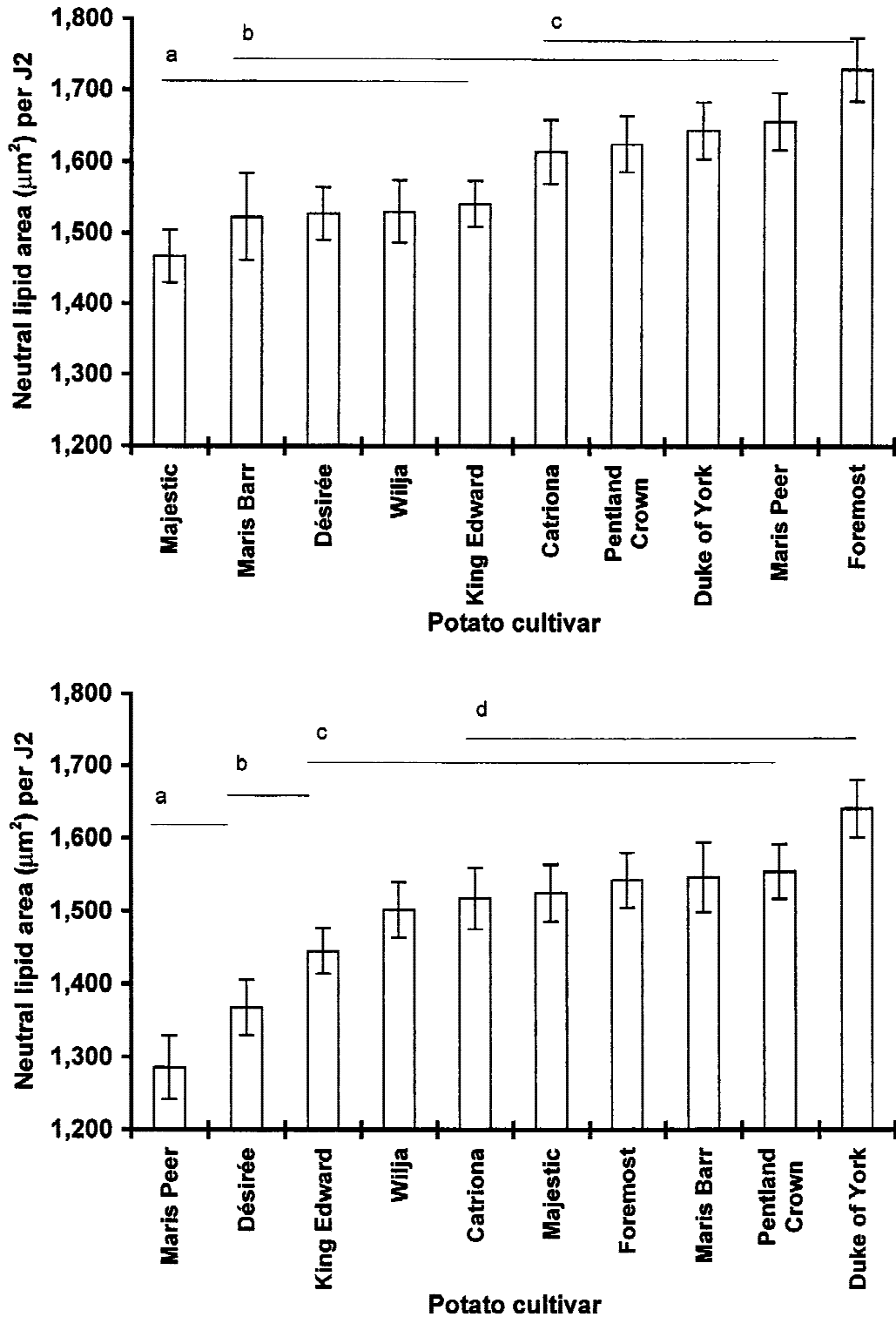


FIG. 2. A) Mean neutral lipid content of second-stage juveniles (J2) of *Globodera rostochiensis* from Bolivia reared on 10 European potato cultivars in pot experiments in Leeds, United Kingdom. B) Mean neutral lipid content of J2 of *G. rostochiensis* from the UK reared on 10 European potato cultivars in pot experiments in Leeds, United Kingdom. Bars represent standard errors of means. Columns grouped by a common horizontal line do not differ ( $P > 0.05$ ).

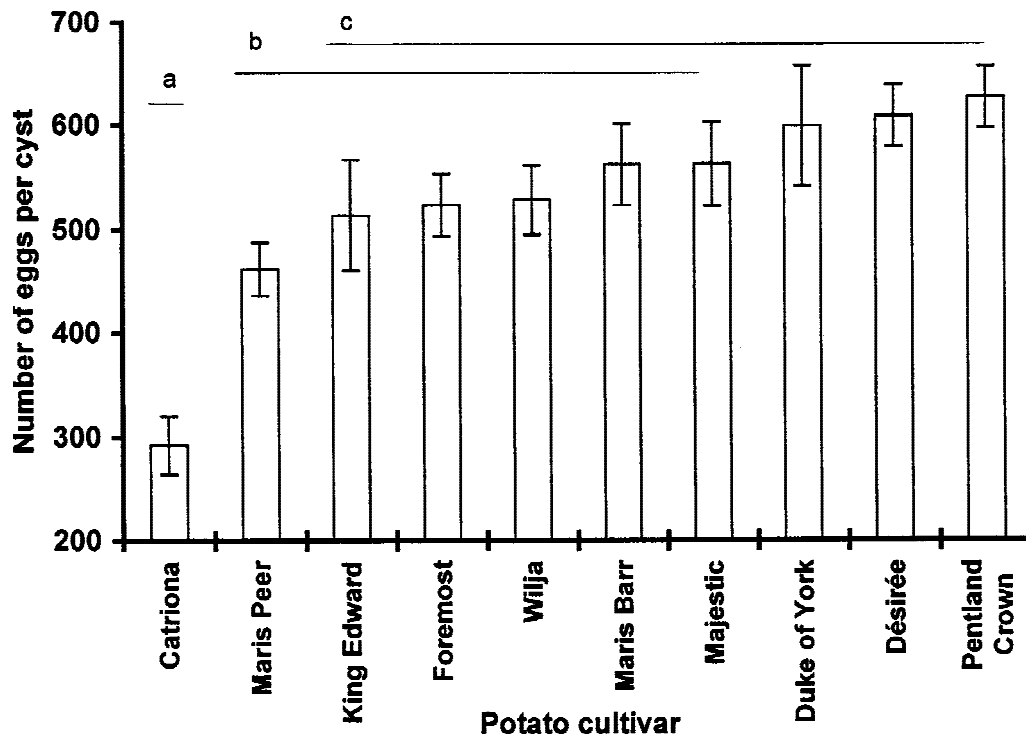


FIG. 3. Mean number of eggs per cyst of Bolivian *Globodera rostochiensis* reared on 10 European potato cultivars in pot experiments in Leeds, United Kingdom. Bars represent standard errors of means. Columns grouped by a common horizontal line do not differ ( $P > 0.05$ ).

content of their progeny (Storey, 1983). Furthermore, females reproducing on cultivars Puca Toralapa and Catriona in the Bolivian and UK experiment, respectively, all provided their progeny with a high neutral lipid content but had low fecundities. The rank correlation was not statistically significant for the two factors ( $P > 0.05$ ), and certain cultivars (e.g., India, Agria, and Waycha) were ranked differently by the two procedures.

In the UK experiment, eight of the ten cultivars yielded a greater lipid content for progeny of Bolivian *G. rostochiensis* than of UK *G. rostochiensis*. A comparison between First Early, Second Early, and Main Crop cultivars found no significant differences among the three subsamples. Cysts of the Bolivian population on nine cultivars contained more than 500 eggs/cyst on average, and some cysts contained more than 1,000 eggs. These numbers were higher than previously reported for *G. rostochiensis* (Brodie et al., 1993; Holz, 1997; Storey, 1983; Whitehead, 1992).

When index scores were combined for both neutral lipid area and number of eggs relative to the overall mean for data from the Bolivian experiment, the scores were much more influenced by variation in egg number than by neutral lipid content (Table 3). The overall combined index range of 18-fold represents a large difference between cultivars. Further work is required to determine if cultivar differences exist for different PCN populations and the diverse geographical zones of Bolivia.

The range of the fatty acids identified in this study,  $C_{14}$  to  $C_{22}$ , is the same as found by others for PCN and *G. tabacum solanacearum*, and is well within the spectrum of most plant-parasitic nematodes (Holz et al., 1997). The 17 fatty acids detected in this study are the same as in UK PCN grown on ssp. *tuberosum* (Holz et al., 1998c).

Results from this study were compared with data published by Holz et al. (1998c) for cysts of British *G. rostochiensis* reared on ssp. *tuberosum* cultivars in the United Kingdom (Table 2). Few differences were found



TABLE 3. Combined index of scores relative to the overall mean for the neutral lipid area and number of eggs for second-stage juveniles of *Globodera rostochiensis* reared on 23 potato cultivars in pots in Cochabamba, Bolivia.

Cultivar	Combined index <sup>a</sup>
Gendarme	0.10
Maria Huanca	0.15
Agria	0.33
Désirée <sup>b</sup>	0.33
Yungay	0.52
Chaposa	0.68
India	0.71
Imilla Negra	0.83
Perla <sup>b</sup>	0.98
Alpha <sup>b</sup>	1.02
Puca Toralapa	1.05
Goyllu	1.07
Waycha	1.20
Lucky	1.23
Colombiana	1.25
Imilla Blanca	1.27
Runa	1.37
Runa Toralapa	1.38
Jaspe	1.47
Sani	1.49
Robusta	1.56
Kallpa Runa	1.62
Kosi	1.83

<sup>a</sup> Combined index is the product of the relative index of the neutral lipid content (1 = mean neutral lipid content of all cultivars put together) multiplied by the relative index of the number of eggs (1 = mean number of eggs of all cultivars put together).

<sup>b</sup> European cultivars, ssp. *tuberosum*. All other cultivars are of Bolivian origin.

among the fatty acid profiles of lipids from Bolivian *G. rostochiensis* cysts reared on cultivars Waycha, Puca Toralapa, and Puna Toralapa in Bolivia. These results are similar to those for ssp. *tuberosum* cultivars (Foremost, Désirée) and a tomato cultivar, in which no major differences in the fatty acid composition of lipids from British *G. rostochiensis* populations reared on these cultivars were found (Holz et al., 1998c).

In the total lipid and neutral lipid fractions, lower relative proportions of C<sub>18</sub> and monounsaturated fatty acids and higher relative percentages of C<sub>20</sub> and polyunsaturated fatty acids were found in Bolivian nematodes than in British nematodes. Greater differences were observed between free fatty acid fractions, wherein relative percentages of the three major fatty acids dif-

fered considerably between Bolivian and British cysts; fewer C<sub>18:1</sub> (20% in Bolivian cysts vs. 28% in British cysts) and C<sub>20:1</sub> (22 vs. 37%) but more C<sub>20:4</sub> (10 vs. 2%) were found in Bolivian cysts.

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