RESEARCHREPORT
PORK SAFETY

Title: Role of gilts in the introduction and transmission of *Salmonella* in swine production systems **NPB #03-026**

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Abstract: Three commercial farrow-to-finish swine production systems in Illinois with a prior history of high prevalence of *Salmonella* infection were evaluated with respect to the degree to which purchased gilts introduce new *Salmonella* genotypes into a herd and acquire *Salmonella* infection from resident pigs. On each farm, one cohort of at least 100 2-week old purchased gilts was assembled, with fecal and pen floor samples collected over time from the gilts and neighboring resident pigs for culture of *Salmonella*. Positive isolates were genotyped using repetitive sequence PCR with REP, BOX, and ERIC primers. The prevalence of shedding *Salmonella* was variable, but generally low (almost always much less than 10%). The analysis of genetic relatedness among isolates showed that, for the most part, closely related isolates were from samples obtained at the same farm visit in the same location, thus indicating limited spread of *Salmonella* beyond the time and place of shedding. Nevertheless, for all 3 production systems, there were several cases of closely related isolates obtained from the pen floors of gilts and resident pigs in the same building or room on the same visit, implicating transmission of *Salmonella* between gilts and resident pigs in both directions. This implicates resident gilts as a potential source of *Salmonella* for resident pigs, as well as a potential conduit for transmission of resident *Salmonella* to other pigs in close proximity. If new genetic strains introduced by purchased gilts are more virulent or have antibiotic resistance, swine and human health could be compromised. These results suggest that separation of pigs from different sources would reduce the introduction and spread of new *Salmonella* genotypes throughout the swine production system.

Introduction: There have been several epidemiologic investigations of *Salmonella* within the environment of swine production systems. In these studies *Salmonella* has been isolated from samples of swine feces, pen floor contents, feed, water, boot swabs, flies, intestinal contents of rodents, and feces of birds and cats. Although numerous sources of *Salmonella* within the environment of the swine production system have been identified, most of the focus on external sources for the introduction of *Salmonella* into a production system has been on the role of feed, with the implication that this is the main conduit for introduction of *Salmonella* into a production system. However, several studies have found little or no *Salmonella* in feed samples. The largest reservoir of *Salmonella* in the swine production ecosystem is undoubtedly swine, because they represent the largest biomass. Likewise, the largest potential source of *Salmonella* for introduction of new infection into the herd is probably females purchased for breeding. Most modern swine production systems replace about 40% of the breeding herd each year, so that for an operation with 1000 sows, there will be 400 gilts introduced into the herd each year from an external source. Transported animals are stressed and are at higher risk for shedding *Salmonella*, which can introduce new infection into the herd. Despite the apparent risk, there has been no published study examining the role of gilts in introducing *Salmonella* into a swine production system.

Objectives: This project was designed to determine the degree to which the introduction of gilts into a swine production system from an external source (1) introduces new *Salmonella* genotypes into the herd; and (2) results in these gilts acquiring infection with resident *Salmonella* genotypes (adding to the *Salmonella* infection load in the herd). Variation in *Salmonella* transmission associated with differences in the degree of gilt isolation (gilts reared on separate site, isolation of gilts on site with other pigs, separation by pen only in nursery) will implicate the role of management in preventing the introduction of new *Salmonella* genotypes into a production system.

Materials & Methods:

(1) Farm Selection and Characteristics

Three commercial swine production facilities in Illinois were investigated. The farms selected had a high prevalence of *Salmonella* based on previous information. Additional inclusion criteria were acquisition of replacement gilts at an age of 2-3 weeks, with at least 100 gilts brought in per shipment. Two-to-three week old replacement gilts were studied because most modern swine production units import replacement stock at this age. A minimum cohort size of 100 gilts was established to identify a sufficient number of *Salmonella* positive isolates to investigate the transmission patterns. All 3 farms housed incoming gilts in total confinement housing units. Gilts were added to each herd at least quarterly and moved to the growing-finishing unit at 6-8 weeks of age and subsequently to the breeding herd at 5-6 months of age or after detection of estrus.

Farm C was a multi-site facility with 3500 sows. Incoming gilts were isolated on a separate site. Farm K was a multi-site facility with 1000 sows. Incoming gilts were mixed within the same nursery room as pigs born on the farm (but in separate pens), as space was available. Farm P was a single-site facility with 600 sows. Incoming gilts were isolated in a separate building. Whereas for farms C and P all stages of production were housed in total confinement, for farm K some gilts at the growing-finishing stage and at

the breeding stage were housed on outdoor lots.

The first visit to each farm was on the day of gilt arrival. A cohort of incoming gilts was selected and ear-tagged for further follow-up. The next visit was approximately one week later. Subsequent visits were within 3 days after each time cohort gilts were moved to another room (usually another building) and then one week later. On each visit, samples were collected from gilts and from an equal number of resident pigs.

On the first and second visits to farm C, samples were collected from resident pigs in an adjacent room of the same nursery unit. On subsequent visits, samples were collected from the cohort and resident pigs kept in the adjacent room in the same finishing unit. In the breeding section, samples were collected from the cohort and other sows in the same pens, because they were mixed in large pens. No follow-up visit was made 1 week after movement of cohort gilts to the breeding unit because the farm was sold to a new owner, who depopulated the herd. Thus, farm C was visited 5 times between May and October 2003 (Table 1).

On the first and second visits to farm K, samples were collected from resident pigs in adjacent pens in the same nursery room in which they were commingled (but separated by pen). In the grower stage cohort pigs were housed in outdoor lots, still separated from resident pigs by pen. Only one visit was made during the growing stage because miscommunication about pig movements resulted in missing the movement of pigs into the grower stage by one week. In the breeding section, samples were collected from the cohort and other sows in the same outdoor pens, because they were mixed together in large pens. This farm was visited 5 times between July 2003 and March 2004 (Table 2).

On the first visit to farm P, samples were collected from resident pigs in the nursery building adjacent to the building where the incoming gilts were housed. No follow-up visit 1 week later was conducted because traffic between buildings was prohibited. On subsequent visits, samples from resident pigs were collected from pens adjacent to the cohort gilts in the growing, finishing and breeding units. Sows were housed in individual pens in the breeding unit, and samples were collected from resident pigs in pens adjacent to the cohort gilts. Due to more frequent movement of pigs, farm P was visited 8 times between November 2003 and July 2004 (Table 3).

(2) Sample Collection

Samples collected included feces from swine and swabs of pen floors and boots. From each pig a 1 gm fecal sample was obtained by digital insertion into the anus, with the collector wearing a latex glove that was changed between each pig sampled. From nursery pigs, feces were obtained using feline fecal loops. Pen floors were sampled to obtain at least 1 gm of floor contents by swabbing using one 4” X 4” cotton gauze square for each pen. Boot samples were obtained by wiping the boots of the data recorder, who remained outside the pen. Upon collection, fecal samples were transferred into tubes containing 9 ml tetrathionate broth with iodine (TTB), and floor or boot samples were placed in tubes with 25 ml TTB, for enrichment of *Salmonella*. TTB tubes were transported to the laboratory in coolers.

(3) Salmonella Culture and Confirmation

Immediately upon return to the laboratory, all samples were incubated at 37 ^{o}C for 48 hr. From each sample 100 µl of TTB broth was transferred to 10 ml Rappaport’s medium (R-10) and incubated for 24 hr at 37 ^{o}C. The enriched samples from R-10 were streaked onto Xylose-Lysine-Tergitol 4 agar (XLT-4) plates and incubated for 24 hr at 37 ^{o}C. Red colonies with black centers were picked and re-streaked onto Brilliant green

agar (BGA) plates. From each XLT-4 plate a maximum of 5 black colonies were picked and transferred to BGA. This allowed identification of multiple genotypes from the same sample. The suspected red non-lactose fermenting *Salmonella* colonies from BGA plates were transferred to Tryptic soy agar (TSA) plates and analyzed using a PCR with primers for the *invA* gene. DNA was extracted by boiling for 5 min the cells obtained from an isolated colony from a TSA plate. All PCR positive isolates were frozen at -80 ^{o}C in Tryptic soy broth (TSB) for further genotypic analysis. *Salmonella* isolates were genotyped using repetitive sequence polymerase chain reaction (rep-PCR) methods with 3 different primers - repetitive extragenic palindromic sequences (REP), BOX, and Entero-bacterial repetitive intergenic consensus (ERIC). The primer sequences for REP-PCR were REP (REP1R-I [5'-IIIICGICGICATCIGGC-3'] and REP2-I [5'-ICGICTTATCIGGCCTAC-3']), ERIC(ERIC1R[5'-ATGTAAGCTCCTGGGGATTCAC-3'] and ERIC2 [5'-AAGTAAGTGACTGGGGTGAGCG-3']) and BOXA, a subunit of the BOX element (BOXA1R[5'-CTACGGCAAGGCGACGCTGACG-3']). REP-PCR produces DNA amplicons that are genomic elements between the primer sites. Resulting DNA amplicons were separated by size by gel electrophoresis.

(4) Data analysis

Data analysis consisted of pairwise comparision of fragmentation patterns to estimate genetic distances between isolates, using the Dice coefficient of fragment matching by size: D_{xy}=1-[2M/(N_x+N_y)], where M= the number of matched size fragments for compared fragmentation patterns, N_x= the number of fragments in the isolate x and N_y= number of fragments in isolate y. A 5% tolerance level for matching was used. Fragment matching data were converted into a distance matrix, with REP, BOX and ERIC data combined into a 3-dimensional distance matrix, using the Euclidean distance. Cluster analysis was performed, using the complete linkage algorithm. Serotyping on all the *Salmonella* positive isolates was performed for farm C to determine whether the same serotypes fell into one cluster and also to compare with the genotype clusters.

Results: For multi-site farm C, 856 samples were collected and cultured for *Salmonella*, 46 of which were positive (Table 4). *Salmonella* positive fecal samples were found only in 22 week old pigs in the breeding barn. None of the samples collected from the nursery at 2-3 weeks of age were positive for *Salmonella*. However, *Salmonella* was detected in the floor samples after the pigs were moved to the growing-finishing section at 9-10 weeks of age. *Salmonella* prevalence was higher for floor samples (45.5%) than for fecal samples (2.2%).

For multi-site farm K, 857 samples were collected and cultured for *Salmonella*, 36 of which were positive (Table 5). On the last 2 visits to the farm, when breeding pigs were sampled, some of the cohort gilts had been moved to a distant breeding site. It was not always possible to differentiate cohort gilts from resident pigs because after housing them on outdoor lots, most of the ear tags had become detached. Thus, an approximately equal number of samples was collected from each site. On farm K no positive fecal samples were obtained from the nursery. Otherwise, *Salmonella* prevalence was variable across visits, class of pigs, and site. *Salmonella* prevalence was higher for floor samples (7.1%) than for fecal samples (3.6%).

For single-site farm P, 2345 samples were collected and cultured for *Salmonella*, 79 of which were positive (Table 6). Salmonella positive samples were found on each visit and there was no association of prevalence with the age of the pigs. *Salmonella* prevalence was higher among the floor samples (5.8%) than the fecal samples (2.6%).

Figures 1-3 show the genetic relatedness among *Salmonella* isolates obtained from each of the 3 farms, as identified by cluster analysis. For all 3 farms, *Salmonella* isolates were differentiated into clusters primarily by spatial location (by building or lot on all farms, also by site on farms C and K), with some secondary separation by visit.

For multi-site farm C, isolates from the 3rd (C13) and 4th (C14) visits, collected from finishing site K, building F (one week apart), formed one large cluster (cluster A) that was distantly linked to isolates from the 5th visit (C15), collected from breeding site W, building F, room 1 (3 months later) (Fig. 1). However, one cluster of isolates from site W, comprised mostly of gilt cohort fecal samples (cluster B), was more closely related to cluster A from site K than either A or B was to cluster C from site W, the latter being comprised of a mixture of isolates from fecal samples of cohort gilts and resident pigs, as well as floor samples from pens in which these animals were mixed. All the positive samples obtained during the 3rd and 4th visits were floor samples. Only 3 were cohort gilt floor samples and the rest were resident pig floor samples. The 2 sub-clusters within the main cluster A, indicated in the Fig. 1 as I and II show isolates from cohort and resident pigs floor sample linked at less than 0.16 linkage distance, suggesting transmission of *Salmonella* between these classes of pigs. Due to a nearly equal number of isolates from each source (gilt, resident pig), it is not possible to attribute a direction to this transmission. Serotyping results showed that clusters formed within 0.4 linkage distance were comprised entirely of the same serotype.

For multi-site farm K, where most samples were collected on the same site, there was separation of the most closely related samples (distance < 0.20) by visit, room, and class of pig (cohort vs. resident), with 3 exceptions noted on Figure 2: T = possible transmission indicated by cluster of gilt fecal and resident floor samples; V = gilt floor sample from visit 3 linked with gilt fecal sample from visit 4; visit 1 had floor samples from pens of both gilts and resident pigs that were genetically similar. (On the day of arrival, it was too early for pig-to-pig transmission to occur, so this probably represents human tracking.)

Figure 3 shows the cluster analysis of genetic relatedness among *Salmonella* isolates from single-site farm P. In most cases the isolates from cohort gilts and resident pigs were in separate clusters. The isolates obtained from the fecal samples of cohort gilt on the day of arrival represent the incoming *Salmonella* genotypes. These were mostly found to be clustered together; however, in some instances they were clustered with isolates from resident gilt feces in subsequent visits. At linkage distances of 0.23 and 0.26, two clusters identified in Figure 3 with black shading had fecal isolates from both resident and cohort pigs. This suggests transmission of *Salmonella* between these classes of pigs. There were many tight clusters with isolates from both fecal and floor samples of cohort gilts and resident pigs, as indicated by the gray shading in Figure 3. Because there are a nearly equal number of isolates from cohort gilt and resident pig samples in this cluster, it is not possible to attribute a direction of transmission.

Discussion: The study conducted here identified *Salmonella* shedding in feces and its presence in pen floor contents in low but variable prevalence, varying according to time, location, age of pigs, and management of gilts, but in no consistent pattern. These results are consistent with previous studies that have found *Salmonella* to be ubiquitous but generally present at low prevalence with occasional outbreaks. With respect to genetic similarity, closely related *Salmonella* isolates, which were from a common source, were in almost all cases from samples collected in a single location on a particular visit, and were from the same class of pigs – either cohort gilts or resident pigs of the same age that had been housed together. Within this context, it is apparent

that in several cases isolates obtained from sampling both cohort gilts and neighboring resident pigs were closely related genetically. This indicates the possibility of occasional transmission of *Salmonella* between these classes of pigs, although it was not possible to infer the predominant direction of transmission.

The degree to which these results generalize to swine production systems in general cannot be readily determined. The farms selected had a history of high *Salmonella* prevalence. Nevertheless, the results of this study present a possible worst case scenario for *Salmonella* transmission and the data still indicate low prevalence and low rates of transmission between different sections of the herd.

The results of this study indicate that incoming gilts do introduce new *Salmonella* onto a farm, adding to the *Salmonella* load on the farm. Although not examined in this study, it is possible that the new genotypes introduced by any incoming gilts may contain characteristics that are harmful in some way to swine and human health, for example by having increased virulence or possessing antibiotic resistance. These *Salmonella* strains could proliferate more rapidly throughout the herd. In order to minimize the introduction and spread of new and dangerous genetic variants of *Salmonella* into a swine production system, it appears that a complete physical separation of cohorts of incoming animals, plus biosecurity measures to prevent *Salmonella* transmission by humans moving between different sections of the herd, would minimize the risk. This can be accomplished readily by maintaining all-in / all-out movement of pigs from the nursery through the finishing stages, but it would also require changes in breeding practices. Separation of the breeding herd into smaller spatially separate units, managed as all-in / all-out facilities by breeding cohorts of the same age and origin, in contrast to the common practice of mixing breeding females in large breeding barns, accompanied by exclusive use of artificial insemination, would reduce the risk of pathogenic *Salmonella* (and other diseases) spreading throughout the herd. This recommendation, of course, needs to be evaluated taking into account other management issues and costs.

Lay Interpretation: Three swine production units in Illinois were examined with respect to the role of purchased gilts in introducing new genetic variants of *Salmonella* into a herd, and the risk of incoming gilts acquiring *Salmonella* infection from resident pigs. On each farm a cohort of at least 100 gilts was followed from day of arrival until the time of introduction into the breeding section of the herd. Fecal and floor samples were obtained from the gilts and from resident pigs in adjacent pens or rooms after each movement of the cohort gilts to a new location, and cultured for *Salmonella*. Polymerase chain reaction (PCR) techniques were used to detect and genetically classify *Salmonella* isolates to identify transmission of the pathogen.

Salmonella was detected at varying times and locations in the production systems examined, although only a small percentage of pigs (usually less than 10%) were shedding *Salmonella* at any time. The close genetic similarity of isolates obtained from samples collected within a room or building on a given farm visit indicates that there was limited mixing of *Salmonella* from different sources on the farm, that is, there is usually a low level of transmission among pigs within different locations on the farm. Close genetic relatedness of some isolates from gilt samples with some isolates from resident pig samples suggests that a small but detectable amount of transmission of *Salmonella* was occurring between gilts and resident pigs that resided in adjacent locations.

Whereas most *Salmonella* infection is subclinical and therefore rarely a problem in reducing productivity, the proliferation of virulent strains of *Salmonella*, some of which may have antibiotic resistance, is a serious concern for swine and human health. Separation of pigs from different sources through all stages of production, even in the breeding herd, as well as biosecurity measures restricting movement of personnel across sections of the herd with pigs from different sources, coupled with exclusive use of artificial insemination, is recommended as the optimal strategy to minimize transmission of different genetic variants of Salmonella across different sections of the herd.

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Table 1: Sample collection at each visit to multi-site farm C

Visit	Date	Cohort Age (Weeks)	Cohort Location	Other Pigs sampled	Notes
1	5/13/2003	2	Nursery	Adjacent room	Day of arrival cohort gilts ear-tagged
2	5/21/2003	3	Nursery	Adjacent room	1week later
3	6/27/2003	8	Finishing	Adjacent room	Day of movement of cohort gilts to finishing building
4	7/5/2003	9	Finishing	Adjacent room	1 week later
5	10/10/2003	22	Breeding	Same room	3 days after movement into breeding building

Table 2: Sample collection at each visit to multi-site farm K

Visit	Date	Cohort Age (Weeks)	Cohort Location	Other Pigs sampled	Notes
1	7/24/2003	2	Nursery	Adjacent pens	Day of arrival cohort gilts ear-tagged
2	7/31/2003	3	Nursery	Adjacent pens	1week later
3	10/5/2003	13	Outdoor Finishing lots	Same & adjacent pens	After moving cohort gilts into outdoor finishing lots
4	3/12/2004	34	Finishing lot & breeding	Same & adjacent pens	After movement of some cohort gilts to breeding
5	3/24/2004	36	Breeding	Adjacent pens	After movement of some cohort pigs into breeding building on distant site

Table 3: Sample collection at each visit to single-site farm P

Visit	Date	Cohort Age (Weeks)	Cohort Location	Other Pigs sampled	Notes
1	11/12/2003	2	Nursery 2	Nursery1 (adjacent building)	Day of arrival cohort gilts ear-tagged
2	1/25/2004	10	Growing stage	Grower & Nursery1	1 day after movement of cohort gilts into grower building
3	2/21/2004	14	Growing stage	Grower	1 day after movement of next batch of gilts into grower building
4	4/18/2004	22	Growing-Finishing	Grower & Finisher	1 day after half of cohort gilts moved to finishing building
5	5/17/2004	26	Finishing	Finisher	1 day after all cohort gilts moved to finishing building
6	6/2/2004	29	Finishing & Gestation	Finisher & Gestation	After 50% of cohort gilts moved to gestation
7	6/14/2004	31	Finishing & Gestation	Finisher & Gestation	After 75% of cohort gilts moved to gestation
8	07/20/04	36	Gestation	Gestation	After 100% of cohort gilts moved to gestation

Table 4: Salmonella prevalence of different sample types by visit for multi-site farm C

		Cohort pig fecal			Cohort pig floor			Resident pig fecal			Resident pig floor			Mixed pig floor		
Visit	**Cohort Age (weeks)**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**
1	2	102	0	0.0	5	0	0.0	0	0	0.0	0	0	0.0			
2	3	99	0	0.0	4	0	0.0	100	0	0.0	10	0	0.0			
3	9	95	0	0.0	2	2	100.0	97	0	0.0	15	11	73.3			
4	10	91	0	0.0	2	1	50.0	97	0	0.0	15	8	53.3			
5	22	67	10	14.9				42	6	14.3				13	8	61.5
Total:		**454**	**10**	**2.2**	**13**	**3**	**23.1**	**336**	**6**	**1.8**	**40**	**19**	**47.5**	**13**	**8**	**61.5**

Table 5: Salmonella prevalence of different sample types by visit for multi-site farm K

		Cohort pig fecal			Cohort pig floor			Resident pig fecal			Resident pig floor		
Visit	**Cohort Age (weeks)**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**%Pos**
1	2	99	0	0.0	7	1	14.3	100	0	0.0	12	3	25.0
2	3	99	0	0.0	7	0	0.0	100	0	0.0	12	0	0.0
3	13	94	5	5.3	1	0	0.0	66	10	15.2	1	1	100.0
Total:		**294**	**5**	**1.7**	**15**	**1**	**0.7**	**266**	**10**	**3.8**	**25**	**4**	**16.0**
		Main site fecal			**Main site floor**			**Distant site fecal**			**Distant site floor**		
Visit	**Cohort Age (weeks)**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**% Pos**	**N**	**Pos**	**%Pos**
4	34	39	2	5.1	11	0	0.0	50	0	0.0	51	0	0.0
5	36	23	7	30.4	22	4	18.2	30	1	3.3	31	2	6.5
Total:		**62**	**9**	**14.5**	**33**	**4**	**12.1**	**80**	**1**	**1.3**	**82**	**2**	**2.4**

Table 6: Salmonella prevalence of different sample types for single-site farm P

		Cohort pig fecal			Cohort pig floor			Resident pig fecal			Resident pig floor		
Visit	**Cohort Age (Weeks)**	**N**	**Positives**	**% Positive**	**N**	**Positives**	**% Positive**	**N**	**Positives**	**% Positive**	**N**	**Positives**	**% Positive**
1	2	120	4	3.3	6	1	16.7	60	3	0.0	6	1	0.0
2	10	120	2	1.7	4	3	75.0	120	6	5.0	10	1	10.0
3	14	114	1	0.9	4	0	0.0	120	8	6.7	6	2	33.3
4	22	106	1	0.9	8	0	0.0	120	2	1.7	8	0	0.0
5	26	113	0	0.0	22	0	0.0	106	2	1.9	50	1	2.0
6	29	120	0	0.0	58	5	8.6	121	1	0.8	65	1	1.5
7	31	90	8	8.9	61	3	4.9	120	2	1.7	89	3	3.4
8	36	93	3	3.2	83	7	8.4	117	2	1.7	105	6	5.7
Total:		**876**	**19**	**2.1**	**246**	**19**	**7.7**	**884**	**26**	**2.9**	**339**	**15**	**4.4**

Figure Legend (Figures 1-3):

The sample identification coding is interpreted as follows:
- First letter code is the farm designation (C, K, P)
- The visit number is written before brackets. Numbers start at 11 (visit 1) to differentiate from a previous study of these farms.
- Designated within brackets for a multi-site farm are (in order, separated by hyphens): site, building, room, pen number, isolate (lower case letter, not separated by hyphen), and sample number; site is omitted for the single site farm.
- After brackets is the stage of production (of the room or lot) and the sample type:
  Stage of production: S- gestation, E- early nursery, L- late nursery, G- grower, F- finisher
  Sample type: F- pig fecal, R- floor.
- After the slash is other information regarding resident and gilt samples:
  R- resident pig or pen, G- cohort gilt or pen, or
  M- mixed floor sample (both gilts and resident pigs in same pen)
  followed cohort ear tag number, if available

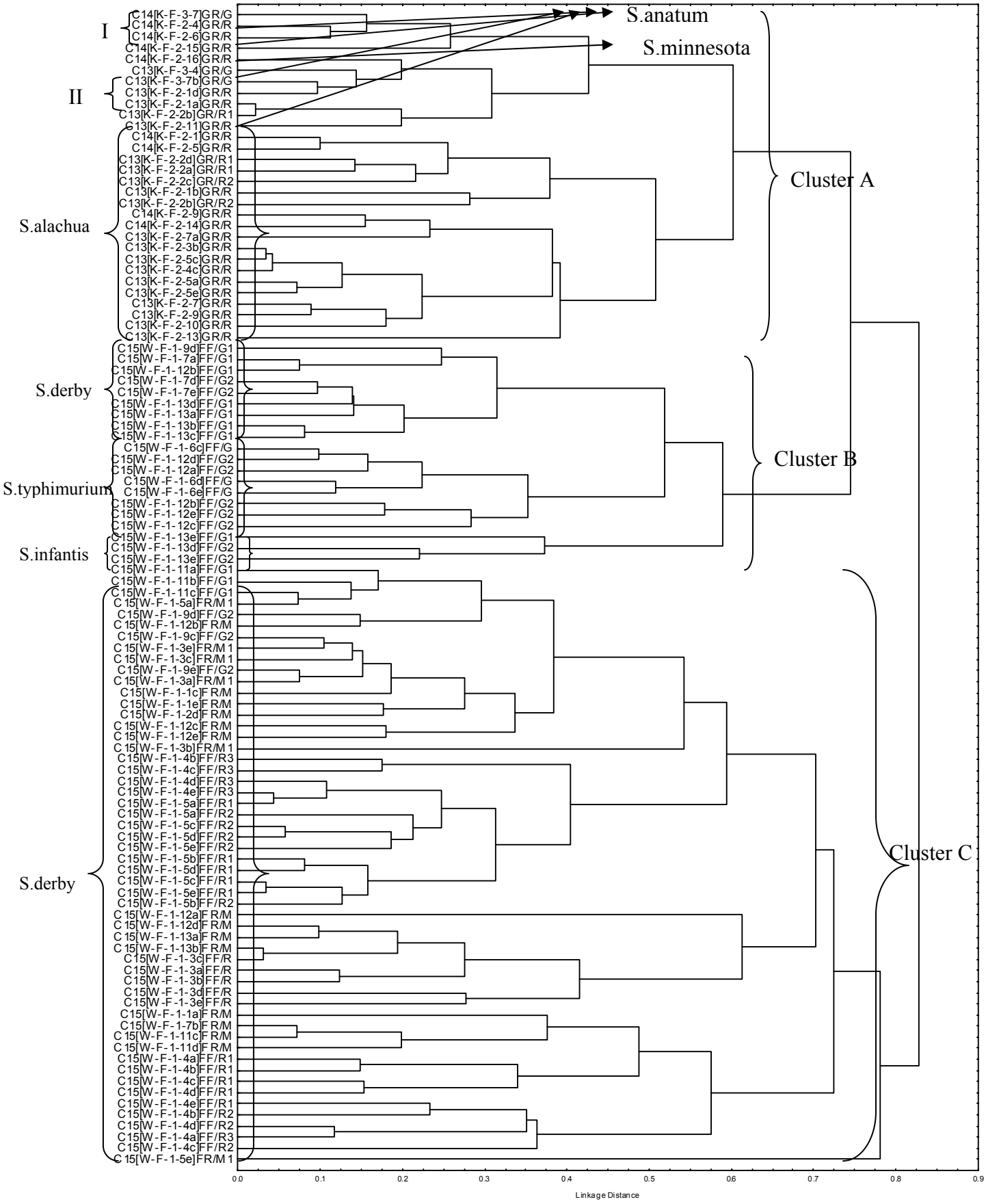


Figure 1. Dendrogram of isolates linked by degree of genetic similarity by cluster analysis for farm C.

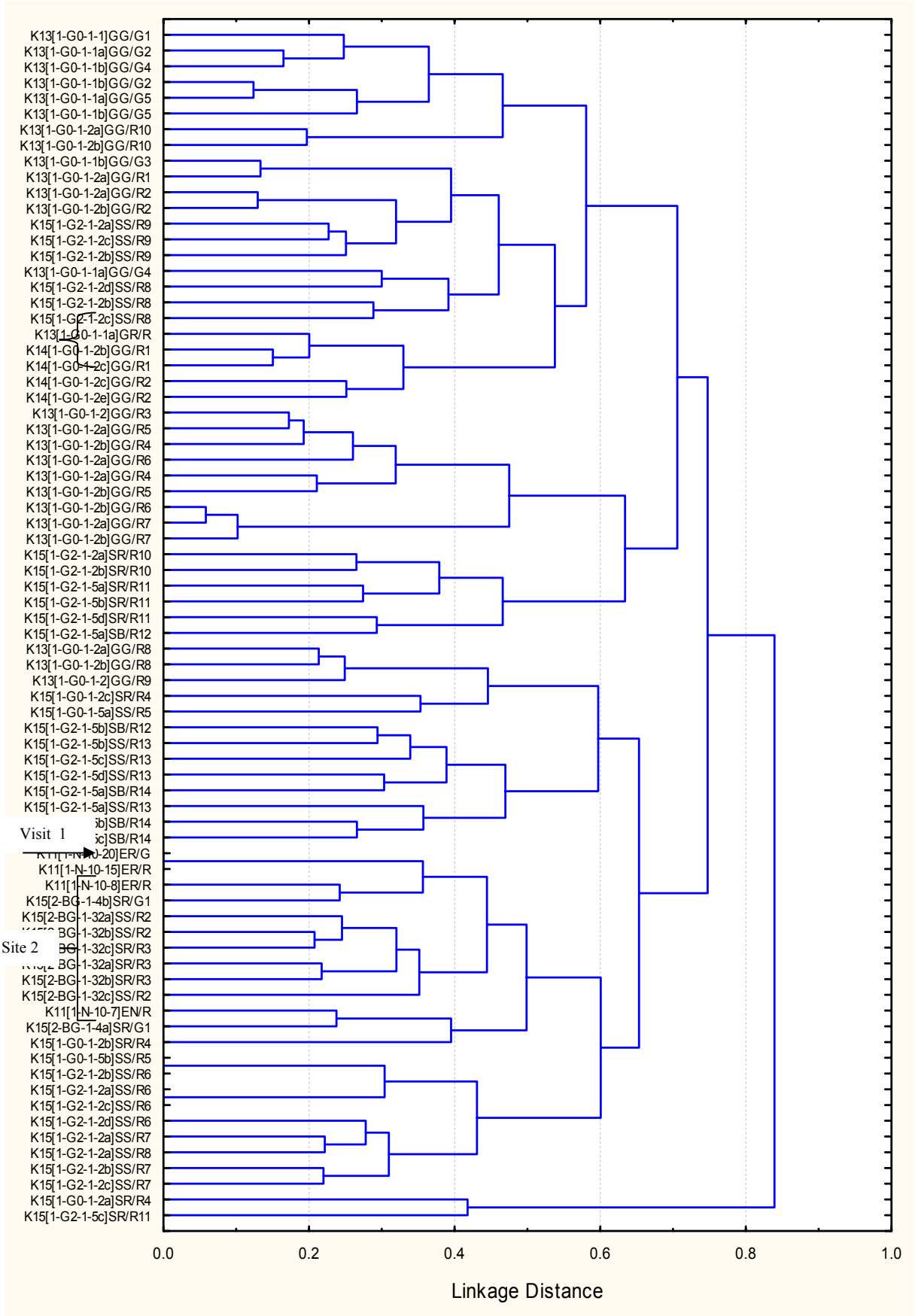
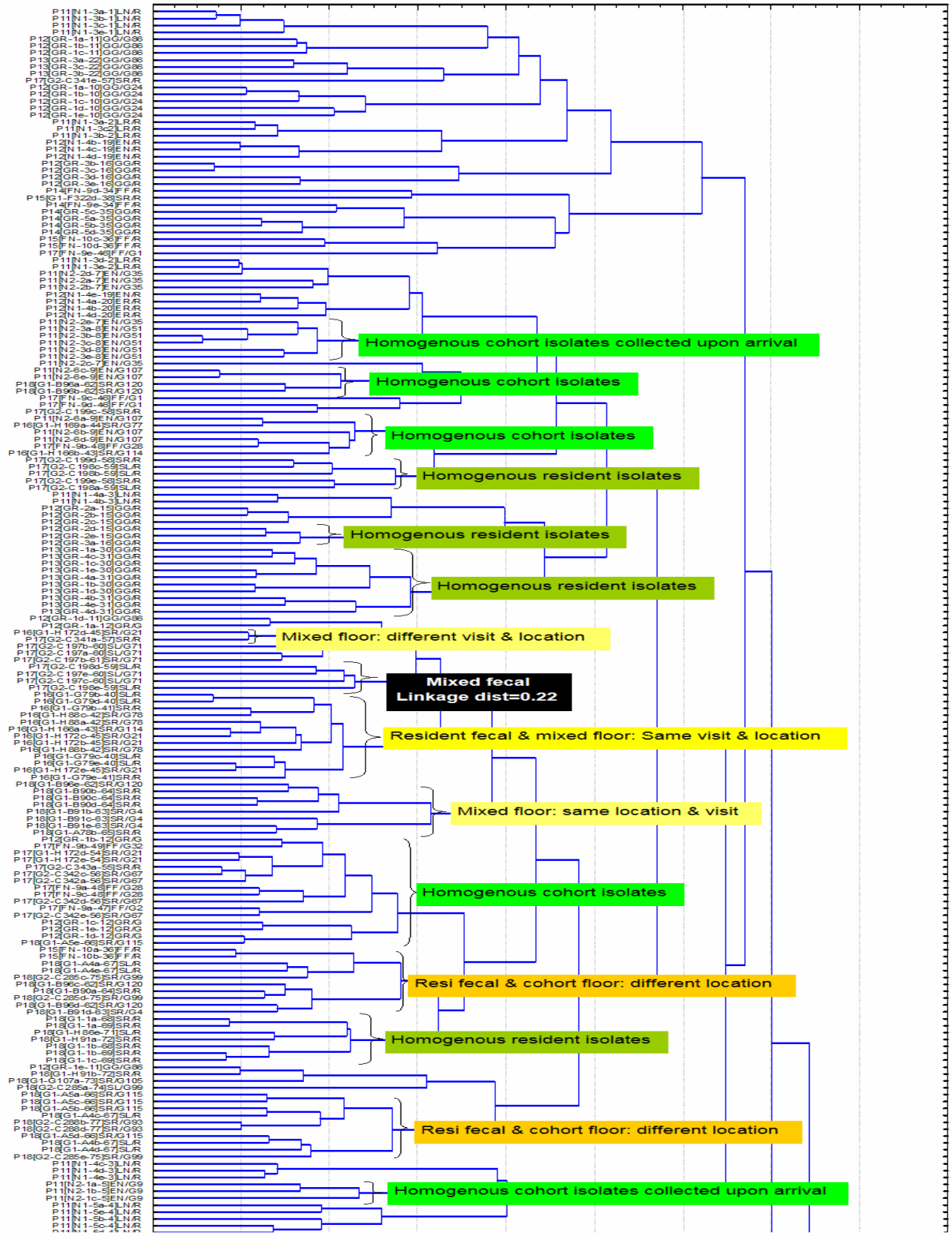


Figure 2. Dendrogram of isolates linked by degree of genetic similarity by cluster analysis for farm K.


Homogenous cohort isolates collected upon arrival
Homogenous cohort isolates
Homogenous cohort isolates
Homogenous resident isolates
Homogenous resident isolates
Homogenous resident isolates
Mixed floor: different visit & location
Mixed fecal
Linkage dist=0.22
Resident fecal & mixed floor: Same visit & location
Mixed floor: same location & visit
Homogenous cohort isolates
Resi fecal & cohort floor: different location
Homogenous resident isolates
Resi fecal & cohort floor: different location
Homogenous cohort isolates collected upon arrival

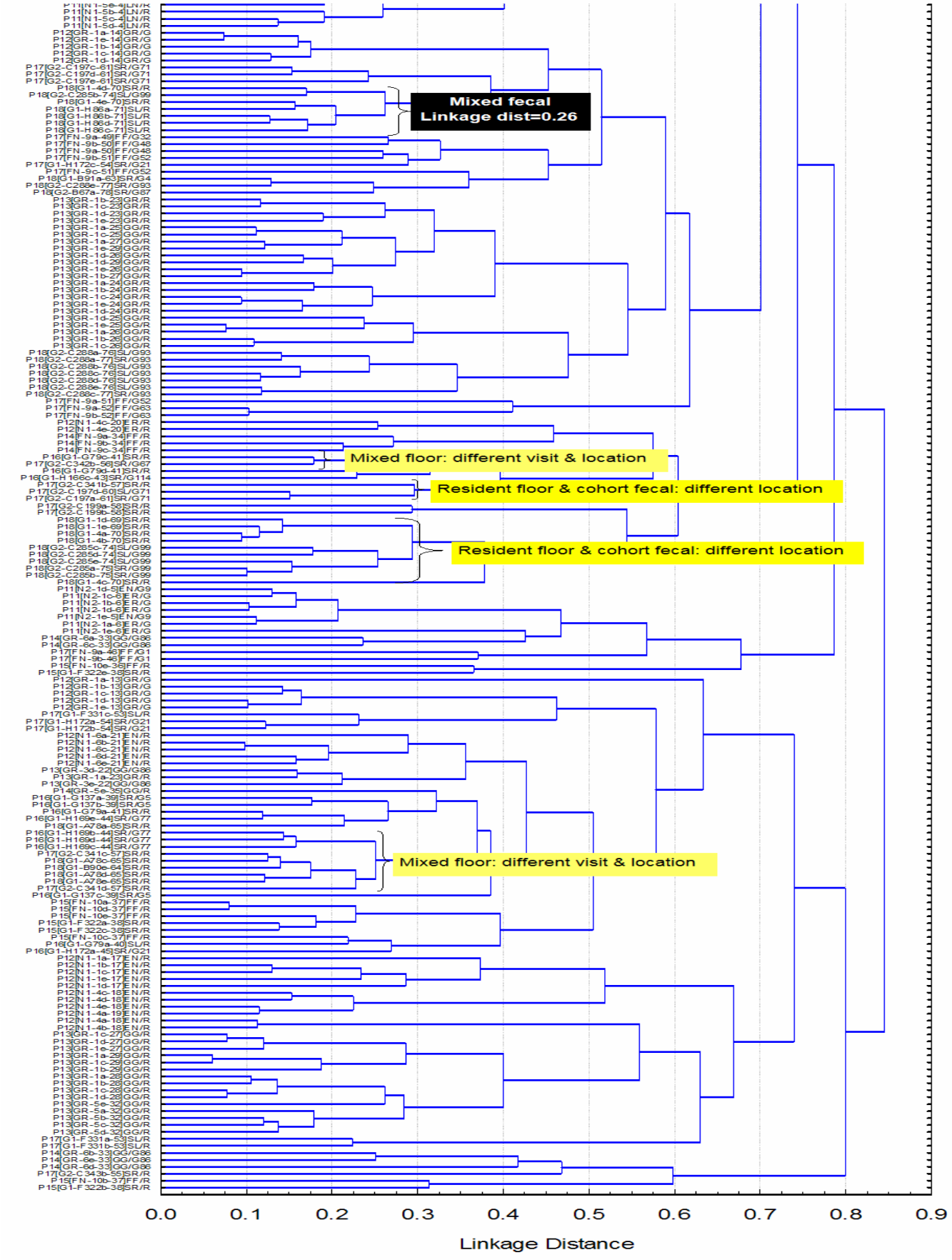


Figure 3. Dendrogram of isolates linked by degree of genetic similarity by cluster analysis for farm P.