The Agr communication system provides a benefit to the populations of *Listeria monocytogenes* in soil

Anne-Laure Vivant¹², Dominique Gamyn¹², Laurent Gal¹²³ and Pascal Piveteau¹²*

¹ Unités Mixtes de Recherche 1347 Agroécologie, Université de Bourgogne, Dijon, France  
² Institut National de la Recherche Agronomique, Unités Mixtes de Recherche 1347 Agroécologie, Dijon, France  
³ AgroSup Dijon, Unités Mixtes de Recherche 1347 Agroécologie, Dijon, France  

*Correspondence:  
Pascal Piveteau, Institut National de la Recherche Agronomique, Unités Mixtes de Recherche 1347 Agroécologie, 17 rue Sully, 21000 Dijon, France  
e-mail: piveteau@u-bourgogne.fr

In this study, we investigated whether the Agr communication system of the pathogenic bacterium *Listeria monocytogenes* was involved in adaptation and competitiveness in soil. Alteration of the ability to communicate, either by deletion of the gene coding the response regulator AgrA (response-negative mutant) or the signal pro-peptide AgrD (signal-negative mutant), did not affect population dynamics in soil that had been sterilized but survival was altered in biotic soil suggesting that the Agr system of *L. monocytogenes* was involved to face the complex soil biotic environment. This was confirmed by a set of co-incubation experiments. The fitness of the response-negative mutant was lower either in the presence or absence of the parental strain but the fitness of the signal-negative mutant depended on the strain with which it was co-incubated. The survival of the signal-negative mutant was higher when co-cultured with the parental strain than when co-cultured with the response-negative mutant. These results showed that the ability to respond to Agr communication provided a benefit to listerial cells to compete. These results might also indicate that in soil, the Agr system controls private goods rather than public goods.

**Keywords:** Agr system, cell communication, competitiveness, fitness, *Listeria monocytogenes*, soil, biotic interaction

**INTRODUCTION**

For the last few decades, communication between bacteria has raised a growing interest. Cell-to-cell communication is based on the synthesis, the diffusion between cells and the perception of signal molecules. The perception of these molecules in the cell's extracellular environment induces the regulation of transcription and eventually adjustment of the physiology of the cell to its surrounding environmental conditions. Various communication systems have been described in the prokaryotic world. They differ according to the type of signal molecules and the machinery used to integrate the signal. To date, the communication systems most studied involve cyclic peptides (AIP), acyl-homoserine lactones (acyl-HSL) or auto-inducer-2 (AI-2) as signal molecules (Miller and Bassler, 2001; Reading and Sperandio, 2006; Atkinson and Williams, 2009).

Several social traits are regulated through cell-to-cell communication. Adhesion, biofilm formation and mobility require functional communication systems in several bacterial species (Labbate et al., 2004; Yarwood et al., 2004; Sturme et al., 2005; Rieu et al., 2007; Boles and Horswill, 2008; Fujii et al., 2008; Jayaraman and Wood, 2008; Riedel et al., 2009; Ray and Visick, 2012; Bowden et al., 2013). Public goods are exo-products as for example, virulence factors, surfactants or antibiotics produced and secreted by bacterial populations. Their production is usually under the control of the spatial distribution and density of cells and is dependent on the characteristics of mass transfer in the environment. For example, in *Staphylococcus aureus* (Morfeldt et al., 1995; Novick and Geisinger, 2008), *Enterococcus faecalis* (Qin et al., 2001; Nakayama et al., 2006), *Clostridium perfringens* (Vidal et al., 2011; Chen and McClane, 2012), *Pseudomonas aeruginosa* (Passador et al., 1993; Pearson et al., 1997), and *Listeria monocytogenes* (Auffray et al., 2003; Riedel et al., 2009), communication systems control the secretion of the virulence factors required for the onset of infection. Moreover, survival mechanisms, such as sporulation, granulose formation, and antibiotic production are also controlled by communication systems in *Clostridium acetobutylicum* (Steiner et al., 2012), *Pseudomonas chlororaphis* (Morohoshi et al., 2013), and *Bacillus subtilis* (Cornella and Grossman, 2005). These communication-dependent coordinated behaviors are examples of cooperation in the microbial world (Keller and Surette, 2006; Diggle et al., 2007). Such a social trait is vulnerable to exploitation by cheaters, these individuals that do not cooperate but gain the benefit from others cooperating (Vélicher, 2003). Cheaters are individuals unable either to respond to the signal or to synthesize it. Cheaters have been isolated from populations of clinical and environmental *P. aeruginosa* (Salunkhe et al., 2005; Heurler et al., 2006). Saving the cost of the production of the signal molecules, of their detection or production of exo-products (Diggle et al., 2007) may give cheaters an advantage and may decrease the value of cooperation (West et al., 2002; Rainey and Rainey, 2003). Experimentally, under controlled environments where access to public goods is required for growth, cheaters are fitter than individuals that cooperate (Rainey and Rainey, 2003; Diggle et al., 2007). Assessing the
value of cooperation in natural settings is required in order to understand why communication and cooperation behaviors have been conserved so far in bacteria.

We tackled this issue with the bacterial model L. monocytogenes as this food-borne pathogen is ubiquitous in nature. It has been isolated from water systems (De Luca et al., 1998; Paillard et al., 2005; Lyautey et al., 2007), vegetation (Welshimer, 1968; Beuchat, 1996), farms (Nightingale et al., 2004; Fox et al., 2009; Latorre et al., 2010; Strawn et al., 2013), food industries (Goulet et al., 1998; Garrido et al., 2009; Serio et al., 2011), and feces of animals (Fenlon, 1985; Iida et al., 1991). It is also found in soil (Welshimer, 1960; Weis and Seeliger, 1975; Locatelli et al., 2013a; Vivant et al., 2013a). A communication system has been characterized in this organism. It is the Agr system that regulates adhesion, biofilm formation (Rieu et al., 2007; Riedel et al., 2009) and infection of mammalian hosts (Autret et al., 2003; Riedel et al., 2009). Four genes, agrBDCA, code the proteins required for Agr communication (Autret et al., 2003; Garmyn et al., 2009). Among them, agrD codes the propeptide AgrD processed into a mature autoinducing peptide (AIP) by AgrB; AgrA, the transcriptional regulator of the two component system AgrC/AgrA, is the response component of the system. Detection of AIP by the sensor AgrC triggers activation of AgrA. In order to investigate whether or not cooperation through communication provided an advantage to populations of L. monocytogenes in complex, natural environments, we compared the behavior of two communication mutants, a signal-negative mutant ΔagrD unable to produce AIP but equipped to sense and respond to AIP, and a response-negative mutant ΔagrA unable to respond to extracellular signal, to the behavior of the parental strain following inoculation in soil.

MATERIALS AND METHODS

BACTERIAL STRAINS AND CULTURE MEDIA

Rifampicin resistant strains were used in this study. The parental strain L. monocytogenes L9 is derived from L. monocytogenes EGD-e (Lemunier et al., 2005). Rifampicin resistant isogenic mutants L. monocytogenes DG125A6 (this study) and L. monocytogenes DG119D9 (this study), respectively are ΔagrA and ΔagrD in-frame deletion mutants (Rieu et al., 2007). Rifampicin resistant strains were isolated on Polymyxin-Acridiflavin-Lithium-Chloride-Cefazidime-Aesclcin-Mannitol agar (PACLM; AES Chemunex, Bruz, France) supplemented with 200 μg.ml⁻¹ rifampicine (Sigma-Aldrich, Saint Quentin Fallavier, France) encapsulating the oxygen content (Lemunier et al., 2005). For each strain, spontaneous RifR mutants were selected by comparing growth rates during planktonic growth and the ability to grow as biofilm in tryptone soy broth (TSB; AES chemunex, Bruz, France) at 25°C without shaking. L. monocytogenes DG125A6 was used as a response-negative mutant and L. monocytogenes DG119D9 as a signal-negative mutant.

A working stock stored at −80°C was used throughout the study. Strains were grown statically at 25°C for 16 h in 5 ml of TSB. Three independent inocula were prepared by inoculating 10 ml of TSB (1% v/v) and incubating statically at 25°C to an O.D₆₀₀ of 0.4. The cultures were then centrifuged at 8000 g for 5 min at room temperature and pellets were suspended in NaCl (0.85%). Cultures were adjusted to a concentration of 2.10⁶ CFU/ml.

SOIL SAMPLES AND SOIL MICRO COSMS PREPARATION

Soil was sampled in a pasture located in Burgundy, France. This sampling site belongs to a country-wide soil sampling network (RMQS) based on a 16 x 16 km systematic grid covering the whole of France (Arrouays et al., 2002). Twenty-five individual core samples of topsoil (0–30 cm) were taken using a sampling design within an area of 20 × 20 m. The core samples were then mixed to obtain a composite sample. The soil sample was then sieved to 5 mm and stored at 4°C. Aliquots of the soil were heat sterilized three times (120°C, 20 min) with a period of 24 h between each autoclave treatment. Fifty g of sterilized and non-sterilized soil were packed in triplicate to constitute sterilized and biotic soil microcosms. Soil’s attributes such as location, composition, chemistry, and land use are stored in the DONESOL database (Grolleau et al., 2004). Briefly, it is a clay soil with neutral pH. Organic carbon and nitrogen content were respectively 35.3 and 3.9 g.kg⁻¹.

SOIL MICRO COSM INOCULATION WITH SINGLE STRAIN AND CO-INOCULATION

Single strain cultivation in biotic and sterilized soil were performed by inoculating a single strain, either L. monocytogenes L9, L. monocytogenes DG125A6 or L. monocytogenes DG119D9, at a concentration of 2.10⁶ CFU/g in 50 g soil microcosms. Microcosms were also co-inoculated with appropriate mixtures from individual cultures to a final ratio of 50:50 (2.10⁶:2.10⁶ CFU/g). The following listerial mixtures were tested: L. monocytogenes L9/L. monocytogenes DG125A6, L. monocytogenes L9/L. monocytogenes DG119D9, and, L. monocytogenes DG125A6/L. monocytogenes DG119D9. Experiments were prepared in triplicates. All inoculated and co-inoculated microcosms were incubated at 25°C in the dark.

ENUMERATION AND DETERMINATION OF LISTERIAL POPULATIONS DYNAMICS

For single-cultures, listerial populations were enumerated by serial plating on Polymyxin-Acridiflawin-Lithium-Chloride-Cefazidime-Aesculcin-Mannitol agar (PACLM; AES Chemunex, Bruz, France) supplemented with 100 μg.l⁻¹ cycloheximide and 100 μg.l⁻¹ rifampicin (Sigma-Aldrich, Saint Quentin Fallavier, France) immediately after inoculation and periodically over a 14-days period for microcosms or over a 48-h period for extracts.

In microcosms inoculated with 50/50 mixtures, the total number of listerial cells was enumerated as described above. The proportion of each of the two strains was determined by strain-specific PCR amplification (described below) from up to 96 colonies collected from the supplemented PALCAM plates.

PCR AMPLIFICATION

DNA template was prepared by transferring each colony in 200 μl of water. Three sets of strain-specific primers were designed to discriminate co-inoculated strains. Two PCR reactions with two of the primer sets were required to discriminate co-inoculated strains. The sequences of the strain-specific primer sets and the
genotype targeted are shown Table 1. PCR amplification was carried out in a final volume of 20 μl containing 2.5 μl of DNA template, 1 μl of dimethyl sulfoxide (DMSO, Sigma-Aldrich, Saint Quentin Fallavier, France), 2 μl of 10X PCR buffer with MgCl₂, 0.16 μl of dNTP mix (25 mM), 1.0 U of Taq polymerase (MP Bio, Illkirch Graffenstaden, France), and a final concentration of 0.6 μM of each primer. The following conditions were specifically determined and used: 95°C for 10 min, 30 cycles of 15 sec at 95°C, 50°C for 1 min and 72°C for 2 min, followed by 7 min at 72°C.

COMPETITIVE INDEX DETERMINATION

For each of the three replicates, the competitive Index (CI) was calculated as follows:

\[
Cl_{tx} = \frac{(\text{CFU}_{\text{mutant}}/\text{CFU}_{\text{parental}})_{tx}/(\text{CFU}_{\text{mutant}}/\text{CFU}_{\text{parental}})_{t0}}
\]

Where \( Cl_{tx} \) is the competitive index at time \( tx \) (\( x = 2 \) days, 4 days, 7 days or 14 days), \( \text{CFU}_{\text{mutant}} \) and \( \text{CFU}_{\text{parental}} \) are the number of Colony Forming Units per gram of soil of the mutant and the parental strains, respectively, at time \( tx \) and at time \( t0 \). A CI score of 1 indicates no fitness difference. A similar calculation was realized for co-cultured listerial mutants.

STATISTICAL ANALYSIS

Patterns of survival of listerial populations were compared by repeated-measures analysis of variance (repeated-measures ANOVA) in both sterilized and biotic microcosms. To estimate whether or not the CI evolved over time, thus to determine whether a strain had a better ability to compete in soil, repeated-measures analysis of variance (repeated-measures ANOVA) was performed.

RESULTS AND DISCUSSION

DYNAMICS OF LISTERIAL POPULATIONS IN SOIL MICRO COSMS

In sterilized soil microcosms, the population of the parental strain \( L. \) monocytogenes L9 increased of over 2 log within the first 2 days of incubation and the population remained stable until the end of the experiment (Figure 1). Inactivation of the Agr system did not affect the dynamics of the mutants' population and no significant differences were observed between growth profiles of the parental strain, the signal-negative \( \Delta agrD \) mutant and the response-negative \( \Delta agrA \) mutant. Similar results were collected during growth in sterilized soil extracts (data not shown). These results confirm previous reports on the ability of \( L. \) monocytogenes to multiply in sterilized soil (Dowe et al., 1997; Moshtaghi et al., 2009; McLaughlin et al., 2011; Piveteau et al., 2011). Moreover, our results suggest that the ability to produce AIP and to respond to the signal is not indispensable for growth of \( L. \) monocytogenes in this specific environment.

When indigenous microflora was not inactivated, in biotic soil microcosms, results were different (Figure 2). First of all, no growth was observed. The population of the parental strain was stable during the first 2 days of incubation thereafter the population declined throughout the duration of the experiment. Furthermore, the behavior of the mutants was significantly different. Indeed, the population of the two mutants declined sharply within the first 2 days of incubation and it was over 1 log lower than that of the parental strain from day 2 to the end of the experiment \( (P < 0.05) \). Differences between mutants were not significant. The results point out to the role of endogenous microbial communities in limiting implantation of \( L. \) monocytogenes in soil. Indeed, inactivation of telluric communities lifts inhibition (Dowe et al., 1997; Locatelli et al., 2013b; Vivant et al., 2013b). Moreover, microbial diversity is critical regarding the ability of soil microbial communities to limit invasión by \( L. \) monocytogenes (Vivant et al., 2013b). Our data strongly suggest that the activity of the Agr communication system is required for optimal survival of \( L. \) monocytogenes in soil. This suggests that the production of signal molecules and/or AgrA-mediated regulation improves the fitness of the populations of \( L. \) monocytogenes in soil. Moreover, production of private or public goods could be involved. In order to figure out if signal sensing in one hand or public goods production in the other hand underpinned the fitness advantage of the parental strain, we followed the fate of populations of the signal-negative and response-negative mutants during co-incubation with the parental strain in soil microcosms.

| Primer set | Oligonucleotide sequence 5′ → 3′ | Genotype targeted |
|------------|-------------------------------|------------------|
|            |                               | \( L. \) monocytogenes: |
|            |                               | L9  | DG125A6 | DG119D9 |
| C10        | CTTCAAAACCGGCATATCAT          | +   | +      | +      |
| C11        | GGAATGTGGCGGAGTTGTT           | +   | -      | +      |
| A19        | AATCCATGTCAGCGGTTCATGTTG     | -   | +      | +      |
| A20        | CTCGAGTAAACTCAAGCTTTTAAAAATT | -   | -      | -      |
| B7         | AGCTACGTCATGAAAAGGTTGCTCTCG  | +   | -      | -      |
| D2         | AAGAATCGGAATTTCTCATG          | +   | -      | -      |

+ amplification, – no amplification.
**COMPARISON OF THE FITNESS OF THE MUTANTS AND PARENTAL STRAINS IN SOIL MICROSCONS**

To determine if alteration of the Agr communication system affected fitness in soil, we measured the survival of the parental strain and of both mutants in sterilized or biotic soil depending on whether they had been cultured with the parental strain, a mutant or as single listerial population. Moreover, Competitive Indexes (CI) of co-cultured listerial strains over a 14-days period in soil microcosms were calculated.

As shown in Figure 3, in sterilized soil microcosms, colonization profiles were similar for all strains whether they had been cultured as a single strain or with a partner. Moreover, variations of the CI were not significant (ANOVA, P > 0.05) (Table 2). This is consistent with the results described above and confirms that in sterilized soil, in the absence of biotic pressure, inactivation of the Agr system does not alter the competitiveness of the mutants. Considering that in sterilized soil, cell density is higher than in biotic soil (about 4 log) and that scavenging of signal molecules is more limited, accumulation of signal molecules is expected.

This suggests that, under these experimental conditions, the AgrA-controlled features may not be essential for growth.

Under biotic conditions, survival of the parental strain (Figure 4A) and the response-negative mutant (Figure 4B) did not vary whatever the co-culture tested. On the opposite, results indicated a significant (ANOVA, P < 0.05) improvement of the signal-negative mutant’s survival when co-cultured with the parental strain but not when co-cultured with the response-negative mutant (Figure 4C). This indicates that the fitness of the signal-mute strain depended of the presence or absence of cells with active Agr systems and that the parental strain provided a benefit to this mutant. In addition to this, CI measurements showed that under biotic conditions, the CI of the response-negative mutant co-incubated with the parental strain significantly (ANOVA, P < 0.05) decreased over time (Table 3). Under these conditions, the parental strain had a significant competitive advantage over the response-negative mutant. The inability to respond to Agr communication was detrimental to the survival of the response-negative mutant. This is supporting the idea that the Agr communication system is important for competitiveness of L. monocytogenes in soil when complex microbial communities are active. When the signal-negative mutant and the parental strain were co-inoculated, the analysis of variance

---

**Table 2 | Competitive Indexes of co-cultured listerial strains over a 14-days period in sterilized soil microcosms.**

| Time (Days) | Response-negative mutant/parental strain | Signal-negative mutant/parental strain | Response-negative mutant/signal-negative mutant |
|------------|------------------------------------------|----------------------------------------|-----------------------------------------------|
| 0          | 1                                        | 1                                      | 1                                             |
| 2          | 1.91                                     | 3.21                                   | 3.74                                          |
| 4          | 1.50                                     | 1.37                                   | 1.03                                          |
| 7          | 1.81                                     | 1.13                                   | 1.63                                          |
| 14         | 1.67                                     | 1.68                                   | 1.12                                          |

* Indicates when the CI significantly differed from the time 0 (repeated-measures ANOVA, Tukey, P < 0.05).
social environment propitious to communication between cells of *L. monocytogenes* even if present in small size populations.

Secondly, these results suggest that cells of the signal-mute population perceive and integrate signals produced by the parental strain into a concerted Agr response that restored the fitness of the signal-mute mutant. Such improvement was not observed with the response-negative mutant suggesting that under these experimental conditions, the Agr communication system regulates intracellular factors (private goods) rather than exo-products (public goods). Production of private goods promotes fitness advantage at the level of the individual cell in the bacterial models *Pseudomonas aeruginosa* and *Bacillus subtilis* (Dandekar et al., 2012; Darch et al., 2012; Oslizlo et al., 2014). Control of private goods by the Agr communication system is supported by results of transcriptomic analyses. Indeed, gene expression profile of the response-negative ΔagrA mutant indicated that deletion of *agrA* resulted in deregulations of amino acids, purine, and pyrimidine synthesis pathways and nitrogen transport (Garmyn et al., 2012). In soil, adaptation of *L. monocytogenes* requires an extensive reprofiling of gene expression (Piveteau et al., 2011) and genes coding proteins involved in cellular processes (transport proteins) and intermediary metabolism (specific pathways for metabolism of carbohydrates) including chitinases and β-glucosidases are upregulated. In the telluric environment where nutrients can be scarce, bacteria must be able to use a large range of carbon and nitrogen sources, for example cellulose and by-products of its hydrolysis (vegetal residues) and chitin (arthropod exoskeleton and cell wall of fungi) polymers largely represented in nature, and to synthesize specific enzymes for their catabolism. The ability of *L. monocytogenes* to acquire and use these energy sources could be critical for its saprophytic life in soil.

Recently, chitin hydrolysis by *L. monocytogenes* was reported to be under the control of the Agr System (Paspaliari et al., 2014). In our experiment, although chitin hydrolysis could generate public goods, we did not evidence any detrimental effect of the presence of mutants to the fitness of the parental strain. Agr mutants did not seem to act as cheaters exploiting the benefit of cooperation under our experimental conditions. In soil, cellular density may be locally inappropriate to gain benefit of cooperation. Others
have shown that induction of public goods can be dominant and mask the benefits of public goods (Dandekar et al., 2012). Considering social traits are vulnerable to cheaters, in populations of L. monocytogenes, Agr mutants should be isolated from environments where Agr communication mediates social traits. However, at the moment, no environmental or clinical isolates of L. monocytogenes have been reported with mutations in agrB-DCA. On the opposite, P. aeruginosa cheaters have been isolated from specific, confined environments where diffusion of signal molecules is low and where the pathogen is able to settle for a long period (Sanzo et al., 2007). Characteristics of the various environments where most isolates of L. monocytogenes have been collected so far are not propitious to the emergence of cheaters.

CONCLUSION
The results reported here give new insights into the role of the Agr communication system in complex natural settings. First of all, the Agr communication system is required for optimal survival of L. monocytogenes in soil; secondly, it provides a benefit to L. monocytogenes populations in soil; thirdly, in the natural environment, production of signal molecules triggers a response in the receiving cells; and fourthly, the Agr system controls private goods. The question of whether the Agr system is a social trait of listerial populations remains to be investigated further. Indeed, the fact that the Agr system controls private goods does not exclude that it also controls public goods in specific habitats of L. monocytogenes.

ACKNOWLEDGMENTS
This work received a grant from the Regional Council of Burgundy (reference PARI AGRALE 11). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

REFERENCES
Arrouays, D., Jolivet, C., Boulonne, L., Bodineau, G., Saby, N., and Grolloe, E. (2002). A new initiative in France: a multi-institutional soil quality monitoring network. Fr. C. R. Agric. 88, 93–103.
Atkinson, S., and Williams, P. (2009). Quorum sensing and social networking in the microbial world. J. R. Soc. Interface 6, 959–978. doi: 10.1098/rsif.2009.0203
Auteert, N., Raynaud, C., Dubail, I., Berche, P., and Charbit, A. (2003). Identification of theagr locus of Listeria monocytogenes: role in bacterial virulence. Infect. Immun. 71, 4463–4471. doi: 10.1128/IAI.71.8.4463-4471.2003
Beuchat, L. R. (1996). Listeria monocytogenes: incidence on vegetables. Food Control 7, 223–228. doi: 10.1016/S0956-7135(96)00039-4
Boles, B. R., and Horswill, A. R. (2008). agr-mediated dispersal of Staphylococcus aureus biofilms. PLoS Pathog. 4:e1000052. doi: 10.1371/journal.ppat.1000052
Bowden, S. D., Hale, N., Chung, J. C. S., Hodgkinson, J. T., Spring, D. R., and West, S. A. (2007). Cooperation and conflict in quorum-sensing bacterial populations. Nature 450, 411–417. doi: 10.1038/nature06279
Dowe, M. I., Jackson, E. D., Mori, I. G., and Bell, C. R. (1997). Listeria monocytogenes survival in soil and incidence in agricultural soils. J. Food Prot. 60, 1201–1207.
Dulla, G., and Lindow, S. E. (2008). Quorum size of Pseudomonas syringae is small and dictated by water availability on the leaf surface. Proc. Natl. Acad. Sci. U.S.A. 105, 3082–3087. doi: 10.1073/pnas.0711723105
Fenlon, D. (1985). Wild birds and sludge as reservoirs of Listeria in the agricultural environment. J. Bacteriol. 59, 537–543.
Fox, E., O’Mahony, T., Clancy, M., Dempsey, R., O’Brien, M., and Jordan, K. (2009). Listeria monocytogenes in the Irish dairy farm environment. J. Food Prot. 72, 1450–1456.
Fujii, T., Ingham, C., Nakayama, J., Beethuyzen, M., Kunuki, R., Molenaar, D., et al. (2008). Two homologous Agr-like quorum-sensing systems cooperatively control adherence, cell morphology, and cell viability properties in Lactobacillus plantarum WCSS1. J. Bacteriol. 190, 7655–7665. doi: 10.1128/JB.01489-07
Garmyn, D., Augagneur, Y., Gal, L., Vivant, A.-L., and Piveteau, P. (2012). Listeria monocytogenes differential transcriptome analysis reveals temperature-dependent Agr regulation and suggests overlaps with other regulons. PLoS ONE 7:e43154. doi: 10.1371/journal.pone.0043154
Garmyn, D., Gal, L., Lemaitre, J.-P., Hartmann, A., and Piveteau, P. (2009). Communication and autoinduction in the species Listeria monocytogenes. Commun. Integr. Biol. 2, 371–374. doi: 10.4161/cib.2.4.8610
Garrido, V., Vitas, A. I., and Garcia-Jalon, I. (2009). Survey of Listeria monocytogenes in ready-to-eat products: prevalence by brands and retail establishments for exposure assessment of listeriosis in Northern Spain. Food Control 20, 986–991. doi: 10.1016/j.foodcont.2008.11.013
Goulet, V., Rocourt, J., Rebire, I., Jacquet, C., Moysé, C., Dehaumont, P., et al. (1998). Listeriosis outbreak associated with the consumption of rillettes in France in 1993. J. Infect. Dis. 177, 155–160. doi: 10.1086/513814
Grolloe, E., Bargeot, L., Chafchafi, A., Hardy, R., Doux, J., Beaudou, A., et al. (2004). Le système d’information national sur les sols: DONESOL et les outils associés. Étude et Gestion des Sols 11, 235–269.
Hense, B. A., Kuttler, C., Mueller, J., Rothballer, M., Hartmann, A., and Kreft, J. U. (2007). Opinion - Does efficiency sensing unify diffusion and quorum sensing? Nut. Rev. Microbiol. 5, 230–239. doi:10.1038/nrmicro1600
Heurlier, K., Denervaud, V., and Haas, D. (2006). Impact of quorum sensing on fitness of Pseudomonas aeruginosa. Int. J. Med. Microbiol. 296, 93–102. doi: 10.1016/j.ijmm.2006.01.043
Iida, T., Kanzaki, M., Maruyama, T., Inoue, S., and Kaneuchi, C. (1991). Prevalence of Listeria monocytogenes in intestinal contents of healthy animals in Japan. J. Vet. Med. Sci. 53, 875–879. doi: 10.1292/jvms.53.873
Jayaraman, A., and Wood, T. K. (2008). Bacterial quorum sensing: signals, circuits, and implications for biofilms and disease. Annu. Rev. Biomed. Eng. 10, 145–167. doi: 10.1146/annurev.bioeng.10.110406.104828
Keller, L., and Surette, M. G. (2006). Communication in bacteria: an ecological and evolutionary perspective. Nature 4, 249–258. doi: 10.1038/nrmicro1383
Labbate, M., Queck, S. Y., Koh, S. K., Rice, S. A., Givskov, M., and Kjelleberg, S. (2004). Quorum sensing-controlled biofilm development in Serratia liquefaciens MG1. J. Bacteriol. 186, 692–698. doi: 10.1128/JB.186.3.692-698.2004
Latorre, A. A., Kessel, J. S. V., Karmali, M. A., Pradhan, A. K., Boor, K. J., et al. (2010). Biofilm in milking equipment on a dairy farm as a potential source of bulk tank milk contamination with Listeria monocytogenes. J. Dairy Sci. 93, 2792–2802. doi: 10.3168/jds.2009-2717
Locatelli, A., Depret, G., Jolivet, C., Henry, S., Dequiedt, S., Piveteau, P., et al. (2013a). Nationwide study of the occurrence of Listeria monocytogenes in 

Frontiers in Cellular and Infection Microbiology www.frontiersin.org November 2014 | Volume 4 | Article 160 | 6
French soils using culture-based and molecular detection methods. J. Microbiol. Methods 93, 242–250. doi: 10.1016/j.mimet.2013.03.017

Locatelli, A., Spor, A., Jolivet, C., Piveteau, P., and Hartmann, A. (2013b). Biotic and abiotic soil properties influence survival of Listeria monocytogenes in soil. PLoS ONE 8:e76991. doi: 10.1371/journal.pone.0076991

Lyautey, E., Lapen, D. R., Wilkes, G., McCleary, K., Pagotto, F., Tyler, K., et al. (2007). Distribution and characteristics of Listeria monocytogenes isolates from surface waters of the South Nation River watershed, Ontario, Canada. Appl. Environ. Microbiol. 73, 5401–5410. doi: 10.1128/AEM.00354-07

McLaughlin, H. P., Casey, P. G., Cotter, J., Gahan, C. G. M., and Hill, C. (2011). LuxU connects quorum sensing to biofilm formation in Listeria monocytogenes. Mol. Microbiol. 71, 1177–1189. doi: 10.1111/j.1365-2958.2008.06589.x

Qin, X., Singh, K. V., Weinstock, G. M., and Murray, B. E. (2001). Characterization of the quorum-sensing system provides a benefit to the populations of Listeria monocytogenes in soil. PLoS ONE 6:e24881. doi: 10.1371/journal.pone.0024881

Rieu, A., Weidmann, S., Garmyn, D., Piveteau, P., and Guzzo, J. (2007). agr system of Listeria monocytogenes EGDe-e: role in adherence and differential expression pattern. Appl. Environ. Microbiol. 73, 6125–6133. doi: 10.1128/AEM.00608-07

Serio, A., Chaves-Lopez, C., and Paparella, A. (2011). Listeria monocytogenes isolated from the smoked salmon industry: growth potential under different environmental conditions. Food Control 22, 2071–2075. doi: 10.1016/j.foodcont.2011.03.010

Steiner, E., Scott, J., Minton, N. P., and Winzer, K. (2012). An agr quorum-sensing system that regulates granule formation and sporulation in Clostridium acetobutylicum. Appl. Environ. Microbiol. 78, 1113–1122. doi: 10.1128/AEM.06376-11

Strawn, L. K., Fortes, E. D., Bihn, E. A., Nightingale, K. G., Gröhn, Y. T., Worobo, R. W., et al. (2013). Landscape and meteorological factors affecting prevalence of three food-borne pathogens in fruit and vegetable farms. Appl. Environ. Microbiol. 79, 588–600. doi: 10.1128/AEM.02491-12

Vivant, A. L., Fortes, E. D., Bihn, E. A., Nightingale, K. G., Gröhn, Y. T., Worobo, R. W., et al. (2013). Landscape and meteorological factors affecting prevalence of three food-borne pathogens in fruit and vegetable farms. Appl. Environ. Microbiol. 79, 588–600. doi: 10.1128/AEM.02491-12

Vivant et al. Agr system and L. monocytogenes behaviors in soil

Vivant, A.-L., Garmyn, D., and Piveteau, P. (2013a). Expression of the agr system in food samples from French urban wastewater treatment plants. Front. Cell. Infect. Microbiol. 3:87. doi: 10.3389/fcimb.2013.00087

Vivant, A.-L., Garmyn, D., and Piveteau, P. (2013a). Listeria monocytogenes, a pathogen down-to-earth. Front. Cell. Infec. Microbiol. 3:87. doi: 10.3389/fcimb.2013.00087

Weis, J., and Seeliger, H. P. R. (1975). Incidence of Listeria monocytogenes in food and soil in Austria. J. Appl. Bacteriol. 42, 541–564. doi: 10.1146/annurev.genet.42.110807.091640

Welshimer, H. J. (1960). Survival of Listeria monocytogenes in soil. J. Bacteriol. 80, 316–320.

Welshimer, H. J. (1968). Isolation of Listeria monocytogenes from vegetation. J. Bacteriol. 95, 300–303.

West, S. A., Pen, I., and Griffin, A. S. (2002). Conflict and cooperation - Cooperation and competition between relatives. Science 296, 72–75. doi: 10.1126/science.1065507

Yarwood, J. M., Bartels, D. J., Volper, E. M., and Greenberg, E. P. (2004). Quorum sensing in Staphylococcus aureus biofilms. J. Bacteriol. 186, 1838–1850. doi: 10.1128/JB.186.6.1838-1850.2004

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 26 August 2014; paper pending published: 24 September 2014; accepted: 17 October 2014; published online: 06 November 2014.

Citation: Vivant A-L, Garmyn D, Gal L and Piveteau P (2014) The Agr communication system provides a benefit to the populations of Listeria monocytogenes in soil. Front. Cell. Infect. Microbiol. 4:160. doi: 10.3389/fcimb.2014.00160

This article was submitted to the journal Frontiers in Cellular and Infection Microbiology.

Copyright © 2014 Vivant, Garmyn, Gal and Piveteau. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) or licensor are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.

Frontiers in Cellular and Infection Microbiology www.frontiersin.org November 2014 | Volume 4 | Article 160 | 7