Review

Cell PRESS

Microbes and Metabolism

Are pathogenic bacteria just looking for food? Metabolism and microbial pathogenesis

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It is interesting to speculate that the evolutionary drive for microbes to develop pathogenic characteristics was to access the nutrient resources that animals provided. Animal environments that pathogens colonize have likely driven the evolution of new bacterial characteristics to maximize these new nutritional opportunities. This review focuses on genomic and functional aspects of pathogen metabolism that allow efficient utilization of nutrient resources provided by animals. Similar to genes encoding specific virulence traits, genes encoding metabolic functions have been horizontally acquired by pathogens to provide a selective advantage in host tissues. Selective advantage in host tissues can also be gained by loss of function mutations that alter metabolic capabilities. Greater understanding of bacterial metabolism within host tissues should be important for increased understanding of host–pathogen interactions and the development of future therapeutic strategies.

Mammals are a rich source of food for bacteria

Animals can be considered an excellent source of nutrients for bacteria. Animal tissues contain a rich diversity of nutrients, including sugars, amino acids and simple nitrogen-containing compounds such as urea and ammonia. This source of nutrients is part of the symbiotic relationship with the microbiota (see Glossary). Pathogens have evolved specific mechanisms to access host nutrients. In this review, we will discuss the intimate evolutionary and functional link between metabolic and bacterial virulence traits.

The interaction of bacterial pathogens with their hosts is distinguished from host–microbiota interactions by resultant host damage [1]. Host–pathogen interactions result in the production and delivery of specific virulence factors that manipulate host cellular processes and cause further responses from the host, including the production of antibacterial factors by the mammalian innate immune system [2,3]. This complex host–pathogen interplay is well described for bacterial virulence secretion systems and innate immune recognition of conserved bacterial molecules [4]. In addition to these exchanges, to grow and replicate bacteria need to extract energy, specifically carbon and nitrogen, from compounds found in this dynamic environment. Bacterial replication is a key factor for pathogen colonization and transmission, hence understanding bacterial metabolism within the host is essential to understand host–pathogen interactions.

Pathogens compete with the resident microbiota

In most habitats, a wide variety of bacteria compete for space and resources. In nutrient-limiting conditions, species that process nutrients more efficiently might outgrow

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Glossary

Carbon catabolite repression: a global regulatory mechanism that inhibits the expression and activities of functions for use of secondary carbon sources when a preferred carbon source is present. This allows bacteria to selectively use substrates from a mixture of different carbon sources.
Catabolic pathway: a series of enzymatic reactions leading to the breakdown of a complex organic molecule to simpler ones with the release of energy.
Gene cluster: any group of two or more genes physically linked in the genome.
Intracellular microorganisms: microorganisms that live and replicate inside a host cell.
Metabolic pathway: a series of enzymatic reactions that converts one biological material to another.
Microbiota (syn. flora): the complete set of microorganisms that inhabit an environment.
Niche: environment in which a species can maintain positive population growth rate.
Pathogenicity island: a discrete genetic unit (with a distinct GC content and a size ranging from 10 to 200 kb) in bacteria, often flanked by direct repeats and often inserted into transfer RNA (tRNA) genes. These islands usually carry genes that contribute to the virulence of the pathogen.
Pseudogene: a gene whose sequence has been mutated and no longer encodes a functional protein.
Quorum sensing (QS): mechanism allowing bacterial cell-to-cell communication and coordination of the expression of various genes in response to a specific diffusable signal.
Stringent response: a bacterial stress response to nutrient limitation signaled by guanosine 5′-(tri)diphosphate,3′-diphosphate [(p)ppGpp]. Accumulation of (p)ppGpp results in downregulation of factors involved in cell growth and upregulation of those required for adaptation to stress, including factors critical for virulence.

others [5]. It is estimated that there are tenfold more bacterial than human cells within the human body. Therefore, in most cases, pathogens have to invade niches that are already occupied by many perfectly adapted resident bacteria. These residents have developed efficient ways to process available nutrients as well as active mechanisms to protect their environment against competing bacterial species [5]. On the skin, the predominant aerobic bacterial species *Staphylococcus epidermidis* produces antimicrobial peptides that are toxic to pathogenic *Staphylococcus aureus* and *Streptococcus pyogenes* [6,7]. In the digestive tract, resident microbiota form a vast heterogeneous microbial ecosystem comprising up to 10^{14} bacteria from more than 400 species. Besides the production of antimicrobial compounds by resident species, the intestinal flora can modulate bone marrow and spleen macrophage cytokine production to promote defense against intracellular microorganisms [8,9]. The vaginal microbiota of healthy, fertile women contains lactic acid generating *Lactobacilli* spp., which maintain the acidic pH of the vagina. The acidic pH as well as hydrogen peroxide production by some *Lactobacilli* spp. can inhibit the growth of many potential colonizers [10]. Therapeutic vaginal re-colonization with hydrogen peroxide producing *Lactobacillus crispatus* prevents recurrent urinary tract infection in susceptible women and reduces the likelihood of urinary tract infection recurrence, which implicates this mechanism as important for vaginal health [11].

In the second section of the paper, we will discuss how pathogens can use original metabolic functions to utilize their niche's resources to overcome competition with resident flora.

Pathogens encounter challenging environments within hosts

Pathogens invading animal hosts colonize diverse changing environments. The pH within the human body is mostly neutral (7.4), but can range from 1.0 in the stomach to 8.0 in the urine. Drastically different environments are also observed as pathogens move from mucosal surfaces deeper into host tissues, such as those observed within the lumen, the multilamellar mucus and the epithelial cells of the stomach [12]. Many environments encountered by bacteria after invasion beyond animal mucosal surfaces are well oxygenated, but the oral cavity, large intestine, female genital tract, abscesses, damaged tissues and the airways of cystic fibrosis patients have areas of low oxygen tension. The level of free iron within mammals is variable (with a mean of 10^{-18} M), but always far below that required for bacterial optimal growth (10^{-6} M), which demands that bacteria rely on their own strategies for scavenging iron [13]. An infected site can be subdivided into numerous physiologically specialized environments that bacteria might encounter or colonize [14]. For example, variable conditions are found between the small intestine, caecum and colon within the intestine [15]. Pathogens might move through multiple diverse environments throughout their life cycle, which could require regulation, coordination and diverse utilization of multiple bacterial metabolic pathways. Bacteria often use metabolic cues to regulate their metabolism and virulence functions.

Dynamic metabolic interactions between hosts and pathogens

Metabolic modulations within host tissues can also be used by pathogens to coordinately regulate virulence factor expression [16]. Carbon catabolite repression triggered in response to carbon source availability influences the virulence of various Gram-negative or Gram-positive pathogens [17,18]. Changes in nutrient supplies, including amino acid and fatty acid limitation, can also trigger the activation of virulence factors via the so-called stringent response through (p)ppGpp [19].

Nutrient availability is obviously not constant in the host. For example, the amount of iron available to bacteria is even lower during infection after the production of host proteins that interact with iron metabolism. First, iron is sequestered by inflammation-induced lactoferrin [20]. Second, lipochelin-2, an antimicrobial protein that captures the bacterial siderophore enterochelin, prevents bacterial iron acquisition. Lipochelin-2 is overproduced in the inflamed intestine in response to enteric pathogens [21]. Many bacterial pathogens sense iron depletion as a signal that they are within a vertebrate host and subsequently modulate the production of virulence factors [22]. The inhibition of diphtheria toxin expression in *Corynebacterium diphtheriae* and Shiga toxin in *Shigella* spp. by the iron-activated global repressors DtxR and Fur, respectively, are among the best studied examples [23,24].

To succeed in a mammal host, bacterial pathogens compete with the resident flora and resist host immune responses. They sense specific environments through variations of nutrient concentrations and subsequently regulate the expression of their virulence factors [16]. In the following section, we illustrate how pathogens utilize metabolic pathways to compete with the resident flora and to cope with harsh environments in the host. From an evolutionary point of view, metabolic genes are then acquired by pathogens in the same way as classical virulence genes. We give examples of metabolic genes linked to pathogenicity and then examine how metabolic constraints can influence the further evolution of pathogens following settlement of a new niche, which could be characteristic of the evolution of virulence (Figure 1).

Metabolic genes help bacterial pathogens colonize new territories

It has been observed time and again that pathogens acquire virulence factors to access new niches [25]. To thrive in these new environments, pathogens also require new metabolic pathways that allow them to exploit available food sources (Figure 1 and Table 1). In these cases, genes that are directly or indirectly implicated in metabolic pathways specific to pathogenic bacteria are absent in their less virulent counterparts. Often these 'metabolic' genes are located on genetic elements (e.g. pathogenicity islands), recently acquired in evolution.

Metabolic pathways can be encoded on pathogenicity islands

The acquisition of genomic islands encoding virulence factors, termed pathogenicity islands, is often essential for the colonization of new host niches. Pathogenicity

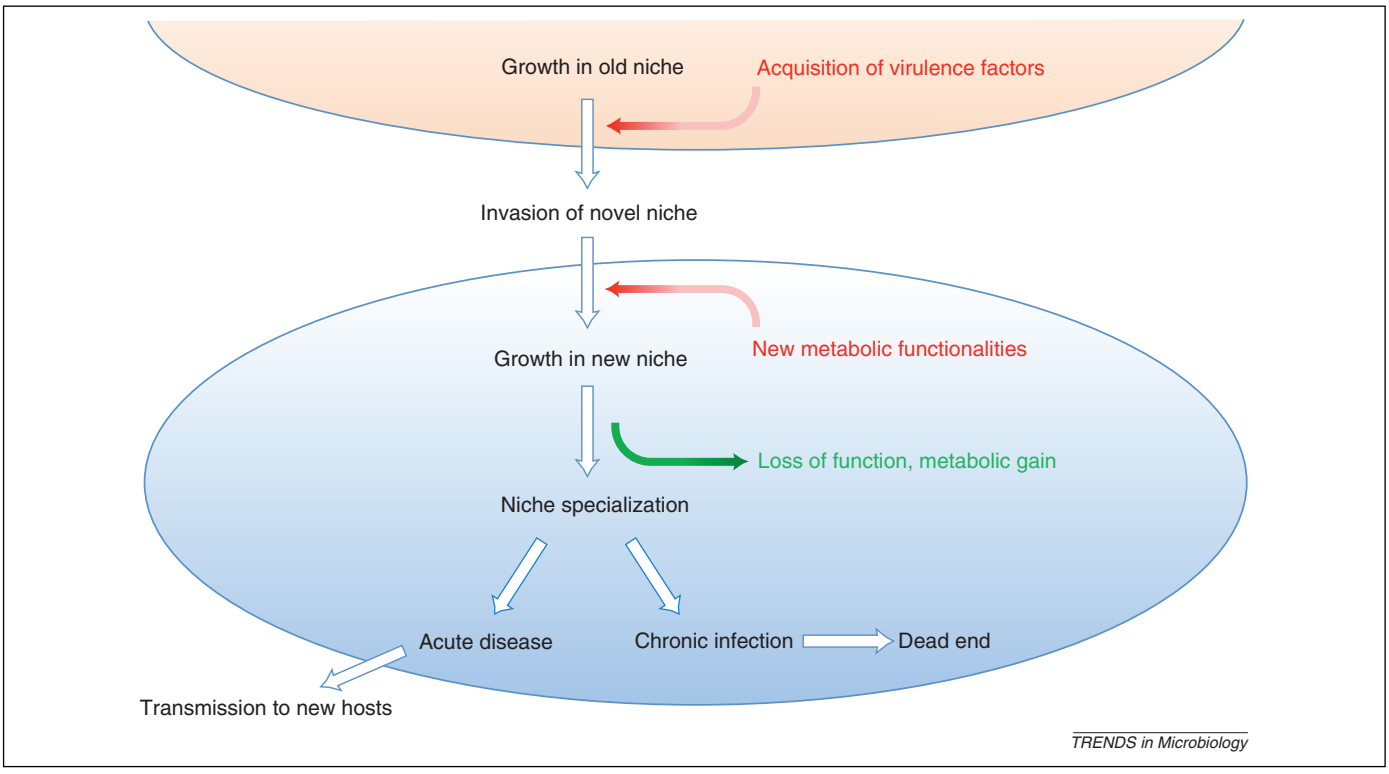

Figure 1. The acquisition of virulence factors along with metabolic capabilities allows bacteria to thrive in a new environment.

islands found in pathogens, but not in their non-pathogenic close relatives, often show evidence of lateral transfer [26]. They sometimes carry genes encoding specific metabolic pathways [25]. Among numerous publications, we took the examples of pathogenic *Salmonella*, *Vibrio* and *Helicobacter* that indicate that these metabolic genes are as important for a successful infection as their classic virulence gene neighbors.

Tetrathionate respiration promotes Salmonella Typhimurium colonization of the intestinal tract

A recent study demonstrated that tetrathionate respiration confers a growth advantage for *Salmonella enterica* subsp. *enterica* serotype Typhimurium in the lumen of the inflamed human intestine [27]. As illustrated in Figure 2, colonic bacteria produce large quantities of highly toxic hydrogen sulfide (H_2S) and the caecal mucosa protects itself by converting H_2S to thiosulfate ($S_2O_3^{2-}$) [27]. Intestinal inflammation induced by *S*. Typhimurium virulence factors (encoded on *Salmonella* Pathogenicity Islands 1 and 2, SPI1 and SPI2, respectively) results in the production of large amounts of nitric oxide radicals and reactive oxygen species in the lumen of the gut. In these conditions, thiosulfate can be oxidized to tetrathionate ($S_4O_6^{2-}$), which selectively inhibits coliforms [27,28]. In contrast to coliforms, *Salmonella* can use tetrathionate to utilize ethanolamine or 1,2-propanediol as carbon sources for anaerobic growth in the intestinal lumen [29]. This process gives *S*. Typhimurium a competitive edge over the gut microbiota, which allows the pathogen to successfully infect the host

Table 1. Examples of acquisition or loss of metabolic-related genes in pathogenic bacteria

Bacterial species	Infected site	Metabolic or regulation pathway acquired or lost	Contribution to virulence	Refs
Gain of function				
Salmonella enterica subsp. *enterica* serotype Typhimurium	Intestine	Cluster *ttrABC, ttrRS* (on SPI2) for utilization of tetrathionate as an electron acceptor	Detoxification, growth	[27,29]
Vibrio cholerae	Intestine	Cluster *nan–nag* (on VPI2) for utilization of sialic acids	Growth	[35]
Helicobacter pylori	Stomach	Gene *nixA* (transferable genomic island) for transport of nickel	Enhanced activity of urease, pH neutralization, chemotactism, inflammation and cell damage	[39–42,44]
Loss of function				
Shigella spp., enteroinvasive *Escherichia coli*	Intestine	Operon *mhp* for propionate degradation	Reduced production of 2-methylcitrate, a gluconeogenesis blocker	[52]
Pseudomonas aeruginosa	CF lung	Mutation in the quorum sensing major regulator *lasR*	Reduced virulence and growth advantage in CF lung	[59,64,67]

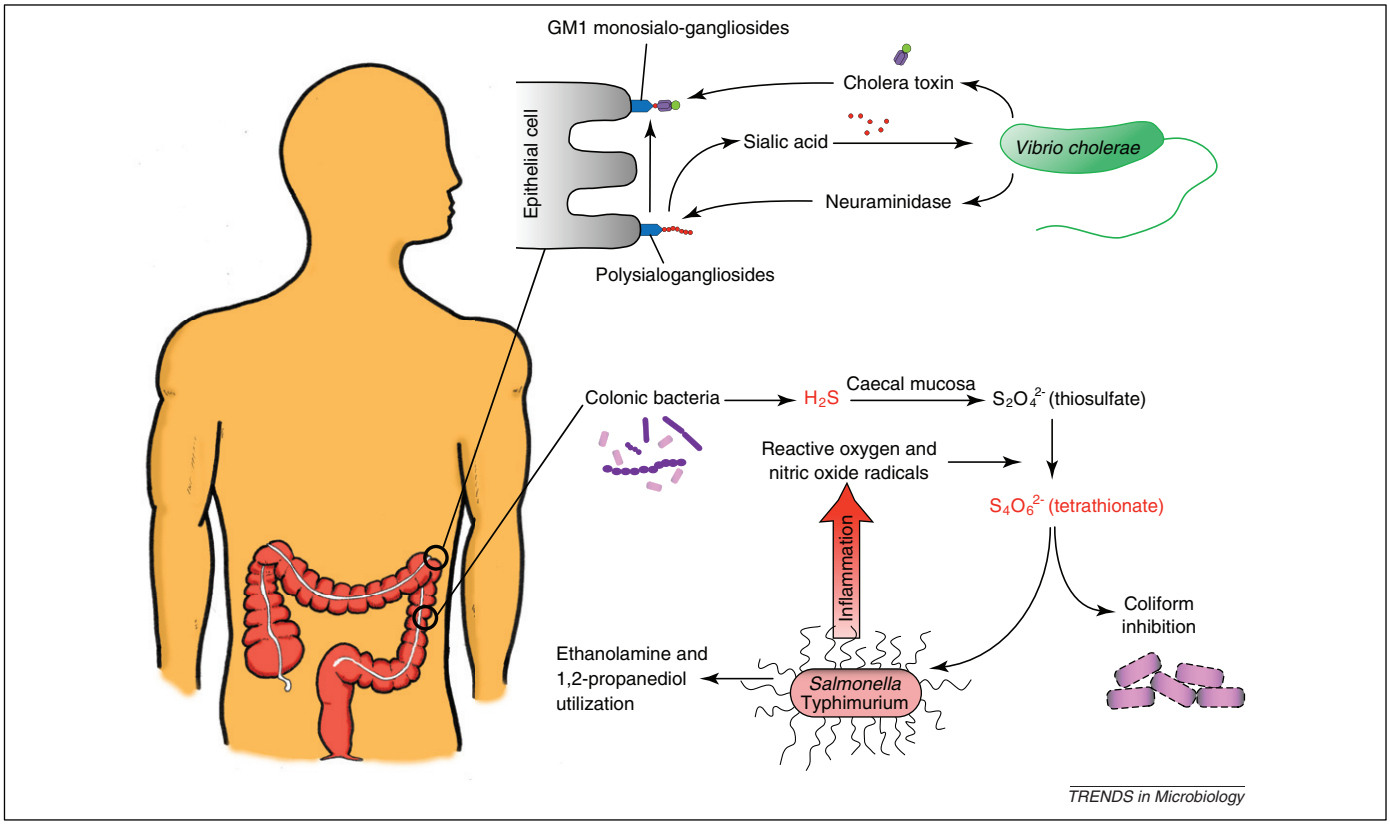

Figure 2. Interplay between bacterial metabolism and virulence pathways in two intestinal pathogens. Sialic acid utilization promotes *Vibrio cholerae* colonization of the intestinal tract [33]. The *Vibrio* pathogenicity island VPI2 is exclusively found in toxinogenic strains. It contains genes encoding a neuraminidase and proteins necessary for transport and catabolism of sialic acids. The neuraminidase converts host cell surface polysialogangliosides to GM1 monoganglioside, the cholera toxin specific receptor, resulting in release of sialic acid. This reaction not only provides an energy source (sialic acid) to *V. cholerae* but also promotes the binding of cholera toxin to host intestinal epithelial cells [33,34]. Tetrathionate respiration promotes *Salmonella enterica* subsp. *enterica* serotype Typhimurium colonization of the intestinal tract. Highly toxic hydrogen sulfide (H_2S) produced by colonic bacteria is converted to less toxic thiosulfate ($S_2O_3^{2-}$) by caecal mucosa. Thiosulfate can be oxidized into tetrathionate ($S_4O_6^{2-}$) by nitric oxide radicals and reactive oxygen species produced by the gut in response to *S.* Typhimurium pathogenesis [27]. Thiosulfate selectively inhibits coliforms [28] but also allows *Salmonella* to utilize ethanolamine or 1,2-propanediol as carbon sources for anaerobic growth in the lumen [29].

and ultimately to achieve transmission to new recipients [30,31].

The five genes responsible for tetrathionate respiration form the *ttrABCRS* cluster located on SPI2, a pathogenicity island critical for the proliferation of *S*. Typhimurium [32]. These genes encode the structural components of the anaerobic tetrathionate reductase (*ttrABC*) and a two-component regulatory system (*ttrRS*) required for the regulation of structural *ttr* genes (Table 1). Because *ttr* seems useful only when inflammatory responses are produced as a result of the production of the SPI1 type III secretion system and its use for invasion, the fixation of SPI2 within the *S*. Typhimurium genome might have been a direct result of the success of strains harboring the *ttrABCRS* cluster. Here, both 'metabolic' and virulence genes are located on the same pathogenicity island (SPI2) and both contribute to intestinal colonization and invasion. This is an interesting example of the physical and functional linkage between virulence and metabolism.

Sialic acid utilization promotes Vibrio cholerae colonization of the intestinal tract

A physical linkage between genes encoding metabolic and virulence functions has also been observed for *Vibrio cholerae* [33]. The *Vibrio* pathogenicity island VPI2 is exclusively found in toxigenic strains and encodes a neuraminidase (from the *nan–nag* cluster) that converts host cell surface polysialogangliosides to GM1 monoganglioside, which specifically binds cholera toxin, by releasing the sialic acid attached to polysialogangliosides (Figure 2). The release of sialic acid from the receptor allows binding of cholera toxin to host intestinal epithelial cells [33,34]. The neuraminidase could therefore specifically provide an energy source (sialic acid) to VPI2-carrying *V. cholerae*. Indeed, in addition to a neuraminidase, the VPI2 pathogenicity island harbors a cluster of genes (*nan–nag*) putatively involved in the scavenging (*nanH*), transport (*dctPQM*) and catabolism (*nanA*, *nanE*, *nanK* and *nagA*) of sialic acids (Figure 2 and Table 1). Inactivation of this catabolic pathway reduced the ability of *V. cholerae* to colonize infant mice, an important animal model for human cholera [35]. Hence the presence of a sialic acid utilization pathway on VPI2 is very important for a successful infection of the human gut by *V. cholerae*.

Interestingly, the *nan* cluster, which allows bacteria to use sialic acid as a carbon source, is found almost exclusively in genomes from bacterial species intimately associated with mammals, most of them pathogens (e.g. *Escherichia coli*, *Shigella* spp., *Salmonella enterica*, *S. aureus* and *Clostridium* spp.). In these species, the *nan* cluster shows extensive signs of horizontal transfer (i.e. incongruent phylogeny and GC content, association with mobile elements and operon structure diversity) [36].

Helicobacter pylori urease activity is enhanced by a nickel transporter

There are few bacteria in the stomach due to the acidic pH, as low as 2, and these consist of mostly transient bacteria swallowed with food and those dislodged from the mouth [6]. For bacteria, urea is an available nutrient in the stomach. It is secreted into the gastric juice through capillary networks beneath the gastric epithelial surface [12]. *Helicobacter pylori* colonizes the stomach and causes gastric lesions such as gastritis, peptic ulcers and gastric cancer [37,38]. Urease activity is an essential factor for stomach colonization by *H. pylori* [12]. *H. pylori* urease is necessary for the bacteria to survive in the low pH found *in vitro*, and urease inhibition abolishes *H. pylori*-related gastric lesions in various animal models [39]. Ammonia generated from urea is a high-quality nitrogen source, neutralizing gastric acidity to give bacteria a neutral microenvironment for their survival and also causing host cell damage and inflammation [39]. Urease activity requires nickel and the high affinity nickel transporter NixA, which contributes to urease activity and full virulence of *H. pylori* (Table 1) [40–42]. Homology search revealed that *nixA* is solely present in the genomes of gastritis-causing *Helicobacter* spp. (e.g. *H. pylori*, *H. felis* and *H. mustelae*) but is missing from the genomes of other *Helicobacter* species that colonize different niches (e.g. *H. hepaticus* and *H. bilis*). Although it is possible that *nixA* was lost in these close relatives, it is more probable that it was acquired by gastritis-causing species subsequent to differentiation between the bacteria that colonize the stomach and those that do not. This is suggested by the fact that the closest homolog to *H. pylori nixA* is found in genomes of *S. aureus*, a species which can cause urinary tract infections, in which optimal urease activity is necessary for virulence [43]. In addition, the genomic region encoding NixA has a significantly lower GC content than the rest of the genome, which suggests horizontal transfer [44]. Hence, *nixA* acquisition might have been crucial in the metabolic adaptation of *H. pylori* to stomach colonization.

Genetic links between new metabolic capacities and virulence factors illustrate that metabolic pathways are acquired as part of the pathogens' evolution towards colonizing new niches with new food sources. Once settled in these new niches, the genome of the pathogen might further evolve to optimize its metabolism through loss of function (Figure 1 and Table 1).

Bacteria can undergo significant metabolic adaptation within diverse host environments

The pathogen life cycle might involve different hosts and host niches with different metabolic nutrient availabilities, which constrains the bacteria to a certain metabolic versatility. This versatility is necessary for a pathogen circulating through different niches in different hosts. For a given pathogen, the metabolic requirement greatly depends on the host infected and the route of inoculation. For example, in *Francisella tularensis*, the tryptophan synthesis pathway is dispensable for intradermal inoculation whereas it is necessary for intranasal inoculation (two natural infection routes), possibly in relation to tryptophan depletion within the inflamed lung [45]. In *Bacillus anthracis*, purine biosynthesis is not required for full virulence in mouse intranasal and rabbit subcutaneous infection models whereas it is required within a mouse intraperitoneal model and in guinea pigs regardless of the administration route [46]. Therefore, the 'metabolic' genes found in a pathogen's genome might be conserved even though they are not necessary once the pathogen reaches its optimal niche. In some circumstances, however, the metabolic capabilities of a pathogen can be altered after loss of function due to mutations, conferring an advantage within a given niche.

Genome reduction is a common adaptation mechanism of pathogens

When pathogens evolve to stably colonize a new niche that offers better nutritional sources, unnecessary or detrimental metabolic pathways can be lost (Figure 1). This hypothesis has been put forth for many pathogens in which genome reduction is ongoing. In particular, specialized intracellular pathogen genomes tend to contain many pseudogenes, which could be due to the abundant nutrient availability in the cell rendering these genes extraneous. For example, in contrast to the intestinal pathogen *S.* Typhimurium, which is largely restricted to the gastrointestinal tract, the systemic pathogens *S.* Typhi and *S.* Paratyphi have lost the tetrathionate respiration pathway genes by independent events, presumably because this pathway does not provide an advantage for extra-intestinal growth [47–49].

It is also possible that a loss of non-essential metabolic functions could contribute to virulence by putting less demand on metabolic pathways. As a proof-of-concept, functional complementation of pseudogenes (metabolic or not) has shown that loss of function could be beneficial for virulence. For example, pathogenic *Shigella* spp. differs from the closely related *E. coli* by the lack of lysine decarboxylase (LDC) activity. Complementing *Shigella* spp. with the gene coding for LDC attenuates its virulence, as a consequence of inhibiting enterotoxin activity by a product of the LDC [50]. In *S. enterica*, the systemic *S.* Typhi differs from the intestinal serovar *S.* Typhimurium by the presence of pseudogenes in *Salmonella* Pathogenicity Island 3 (SPI3). Complementation of *S.* Typhi with the SPI3 genes (with unknown functions) from *S.* Typhimurium dramatically reduced the invasion of monocytes [51]. Comparative genomics also reveals repeated loss of metabolic pathways in some species. *Shigella* spp. and enteroinvasive *E. coli* arose from distinct *Escherichia* ancestors, acquiring the capacity to invade epithelial cells. The genomes of these species underwent substantial reduction, and multiple metabolic pathways were lost. Among them, a propionate degradation pathway was specifically deleted from the genome of these enteroinvasive pathogens although it is found in all other *E. coli* genomes [52]. The propionate degradation pathway was shown to produce 2-methylcitrate, which blocks the activity of the gluconeogenic enzyme fructose-1,6-bisphosphatase, a critical component of virulence for pathogenic microorganisms [53–55]. Hence, these pathways could potentially be selected against because of their non-essentiality and mildly detrimental effect during the intracellular life cycle.

Gene loss in Pseudomonas aeruginosa that infect airways of patients with cystic fibrosis

In chronic infection, pathogens are prone to reduce their genome, which can give them a metabolic edge. One of the best examples is *Pseudomonas aeruginosa*, which infects airways of cystic fibrosis (CF) patients. CF is a genetic disorder resulting from mutation in the CF transmembrane conductor regulator (CFTR) gene, which encodes a chloride channel in secretory epithelia. It results in the accumulation of a thick mucus within the lung, which allows colonization and growth of multiple opportunistic pathogens [56]. Infection with the Gram-negative opportunistic bacterium *P. aeruginosa,* which can colonize its host for decades, is associated with a significantly poorer respiratory outcome [57,58]. It has long been known that a major characteristic of *P. aeruginosa* isolated from these chronically infected airways is the loss of a variety of metabolic pathways [59]. This is probably related to the large amount of available nutrients (e.g. amino acids, nitrate and other nitrogen species) in the airway environment, as compared to soil and water, the normal habitat of *P. aeruginosa* [59–63]. *P. aeruginosa* has been shown to adapt to these particular conditions while colonizing the CF lung [59]. The expression of several acute virulence factors important to initial colonization in mouse models of infection, such as elastase and pyocyanin, are regulated by the quorum sensing (QS) systems [64]. Interestingly, growth in the rich environment of the CF lung results in loss of *lasR*, which encodes the major QS transcriptional regulator [65]. The mechanisms underlying the selection of *lasR* mutations are unknown but could be driven by nutrient availability within the CF airway. Indeed, *lasR* mutants have a growth advantage when cultured on particular amino acids, in part due to an increased expression of the catabolic pathway regulator *cbrB*, and utilize more nitrate compared with their isogenic parents [66,67]. Because LasR also controls virulence factor production, the increased nutrient availability within the CF airway could select for less virulent bacteria better adapted to chronic infections.

Such a balance between self preservation and nutritional competence (called 'SPANC' by Ferenci *et al.*) is observed in many other pathogenic bacteria [68]. When a pathogen establishes itself into a niche where nutritional competence brings more benefits than its virulence function, the balance is altered and mutational adaptations occur that change the regulation of virulence and metabolic genes.

These examples illustrate how a loss of function in a pathogen can impact its metabolism and virulence. The prevalence of such mutations within species depends on the impact of such adaptations on bacterial life cycles (Figure 1).

The future of studying the role of bacterial metabolism in microbial pathogens

Since pathogenic microbes rapidly develop resistance against existing antibiotics, new anti-infective strategies are needed to keep ahead of the inevitable resistance that accompanies antimicrobial use. Since metabolism is a prerequisite for virulence, such pathways could potentially be good targets for anti-microbial therapies. Bacterial *in vivo* metabolism is one of the most fundamental aspects of virulence of pathogenic bacteria yet our understanding of it is relatively limited.

Our current view of bacterial metabolism in host tissues is largely derived from the investigation of deletion mutants within inbred mouse models and the transcriptional and proteomic profiling of bacteria obtained during infection of animal models. Some of the current data derived from these *in vivo* high-throughput screens aimed at identifying metabolic genes essential for infection are inconsistent with each other: the essential genes depend on the model system used, the type of infection and the route of inoculation. It also underlines that high-throughput screen data should be considered with some caution because their results might not necessarily be generalized to other infection models, especially when it comes to metabolic genes [69].

Although comparative genomics should identify new pathways unique to specific pathogens or associated with virulence genes, new strategies for the analysis of the importance of specific metabolic pathways in host tissues must be developed. Particularly promising is the use of radioisotopes to elucidate carbon and nitrogen sources used by pathogens over the course of an infection [70,71].

The role of metabolism in virulence should be increasingly recognized as a priority equivalent to studying classical virulence factors (Figure 1). Metabolic genes are often identified by global studies attempting to identify virulence genes but are rarely further investigated. Missing metabolic pathways in genomes from pathogens have also often been dismissed as the result of lack of selection due to the nutrient-rich environment in the host. However, new data suggest that their loss might be advantageous for virulence [50,51]. Future studies could focus on those bacterial metabolic genes in which genome analysis suggests involvement in host–pathogen interactions.

To date, antimicrobial therapies based on interference with bacterial metabolism have been largely limited to folate metabolism. Recently, bacterial virulence factors have begun to be reconsidered as possible therapeutic targets. The idea that inhibition of bacterial virulence characteristics could be used therapeutically is attractive because it would not require that antimicrobials kill bacteria and hence reduce selection for resistance. Some work is ongoing in this area. Bacterial urease is a potential metabolic target for the development of new therapeutics to manage gastric and urinary tract infections. Hydroxamic acids or phosphoro-diamidates strongly inhibit ureases *in vitro* and one compound (acetohydroxamic acid) is already available in some countries as adjunctive therapy in patients with chronic urea-splitting urinary infection [39]. In the same vein, interruption of iron trafficking is a plausible but still largely unproven means of clinically controlling pathogens [72,73].

Concluding remarks

In conclusion, a greater understanding of bacterial metabolism specific to infection of multicellular organisms should ultimately lead to a deeper understanding of bacterial pathogenesis as well as host metabolism. It might also identify new strategies for antimicrobial chemotherapy that would be specific to pathogens.

Acknowledgments

L.R. and S.I.M. were supported by the National Institute of Allergy and Infectious Diseases (NIH grants 10265SUB and U54AI057141) and by the Cystic Fibrosis Foundation (Grant R565-CR07). D.H. was supported by a grant from the European Community (7th PCRD, PIOF-GA-2009-235009). We thank the members of the Miller lab for fruitful discussions and Rodolphe 'Rodho' Grandviennot for the drawing of the cover and in Figure 1.

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