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Murine infection models for the investigation of *Campylobacter jejuni* – host interactions and pathogenicity

Murine tierexperimentelle Infektionsmodelle für die Analyse der Wirtsinteraktionen und die Pathogenität von Campylobacter jejuni

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Summary

Despite the socioeconomic burden of campylobacteriosis, our insights into the molecular mechanisms underlying *Campylobacter* (*C.*) *jejuni*-induced intestinal immunopathogenesis are limited. The absence and the ban of convenient murine infection models caused fundamental restrictions in *Campylobacter* research. The development of novel and modified murine infection models in the last years has greatly contributed to our knowledge in *C. jejuni* – host interactions and pathogenici ty. Novel findings revealed that the colonization resistance of mice against *C. jejuni* infection can be overcome by modification of the intestinal microbiota. In particular *C. jejuni* infected infant mice harbouring a conventional microbiota and Interleukin-10-deficient mice rendered gnotobiotic by antibiotic treatment develop lipooligosaccharide mediated inflammation and specific T-cell responses – both key features of campylobacteriosis in humans. This short review focuses on the major progress in this growing research field and intends to summarize some of the most important findings.

Keywords: *Campylobacter*, animal models, colonization resistance, microbiota

Zusammenfassung

Trotz der wachsenden Bedeutung von *Campylobacter* (*C.*) *jejuni* als Enteritiserreger weltweit ist unser Verständnis der molekularen Pathogenitätsmechanismen der Campylobacteriose eingeschränkt. Die grundsätzliche Entscheidung, die Maus als Tiermodell für die *C. jejuni*-Infektion nicht zu verwenden, hat zu einer fundamentalen Restriktion der Grundlagenforschung und zu einem Verzug im Erkenntnisgewinn geführt. Daraus resultiert ein dringlicher Forschungsbedarf, der sich auf die Mechanismen der Wirtsinteraktion bezieht und deshalb auf Tiermodelle in der Maus besonders angewiesen ist. In diesem Übersichtsartikel möchten wir über die neuen, innovativen und aktuellen Veränderungen im Forschungsfeld der murinen Infektionsmodelle für *C. jejuni* berichten. Neue Erkenntnisse belegen, dass die Kolonisationsresistenz von Mäusen gegen *C. jejuni* durch Modifikation oder Eradikation der intestinalen Mikrobiota aufgehoben werden kann, und dass entsprechend modifizierte murine Infektionsmodelle für die Aufklärung der Pathogenitätsmechanismen von *C. jejuni* besonders geeignet sind. So zeigen junge Mäuse mit einer konventionellen Mikrobiota und Interleukin-10 defiziente Tiere, die durch Antibiose gnotobiotisch gemacht worden waren, sowohl den Verlauf als auch die grundlegenden immunologischen Eigenschaften der Campylobacteriose beim Menschen. Dies wird am Beispiel der durch Toll-like-Rezeptor 4 vermittelten Verstärkung der Entzündung durch Lipooligosaccride und durch spezifische T-Zellantworten in gnotobiotischen und Interleukin-10 defizienten Mäusen besonders hervorgehoben.

Schlüsselwörter: *Campylobacter*, Tiermodelle, Kolonisationsresistenz, Mikrobiota

Murine infection models in *C. jejuni* research – a long and winding road

Investigations of molecular mechanisms underlying human campylobacteriosis were hampered and needlessly delayed for decades due to the lack of appropriate animal models (Alter et al., 2011; Kist and Bereswill, 2001; Bereswill und Kist, 2002, 2003; Dorrell and Wren, 2007; Young et al., 2000). Piglets, ferrets, primates, and chicken have been used as infection models with varying but mostly poor success. Mice are highly convenient for the investigation of many pathogenic bacteria. However, rodents display a strong colonization resistance against *C. jejuni* caused by the normal murine gut microbiota (reviewed by Dorrell and Wren, 2007; Young et al., 2000). In consequence, conventional mouse infection models for human campylobacteriosis suffered from sporadic colonization, absence of symptoms and poor reproducibility. These shortcomings led to a ban of murine *C. jejuni* infection models at the end of the 20th century (Dorrell and Wren, 2007; Young et al., 2000). Therefore, it is still unclear how this important pathogen causes intestinal inflammation and systemic complications such as Guillain-Barré syndrome (GBS) in humans. In order to increase our knowledge about the molecular mechanisms underlying infection, pathogenesis of inflammation, and long-term colonization of *C. jejuni* novel mouse models have been developed recently. *C. jejuni*-infected mice with a modified microbiota composition and/or immune system developed key symptoms of campylobacteriosis in humans. The latter approaches included NF-kB-deficient mice (Fox et al., 2004), mice lacking the innate signalling adapter protein MyD88 or SCID mice that have been successfully used for the study of immune responses during *C. jejuni* infection (reviewed by Hodgeson et al., 1998; Dorrell and Wren, 2007). In particular experiments with *C. jejuni*-infected MyD88-deficient mice revealed ground-breaking results providing the first evidence that the innate immune system is essentially involved in the initiation and progress of *C. jejuni* mediated enteritis. However, the use of these highly immunocompromized mice as reliable models for the study of pathogens in general is limited by the development of severe clinical conditions even without *C. jejuni*-infection.

The use of germfree mice for campylobacteriosis research

Recent results confirmed that the strict colonization resistance of mice against *C. jejuni* is caused by the conventional murine microbiota. This was proven experimentally earlier by the effective colonization of *C. jejuni* in mice with a defined limited gut microbiota (Chang and Miller, 2006), in Streptomicin-treated mice, and in isolator-raised germfree mice (Yrios and Balish, 1986a, b, c; Youssef et al., 1987; Jesudason et al., 1989). In contrast the vast majority of mice with a conventional microbiota expelled the pathogen efficiently within a day. Moreover, results of these ground-breaking experiments demonstrated that in germfree mice *C. jejuni* not only colonizes the entire gastrointestinal (GI) tract but also induces clinical signs of disease. Immunopathogenesis induced in the GI tract included granulocytic infiltrates, bloody diarrhea, and *C. jejuni* infection was reproducibly accompanied by humoral immune responses. This led to the insight that the murine intestinal microbiota is responsible for sporadic pathogenic colonization and thus for the shortcomings of mouse models including the lack of standardization of results in different laboratories (Dorrell and Wren, 2007).

In order to overcome experimental limitations due to physiological colonization resistance (Young et al., 2000) various researchers including ourselves have focused on the development of novel murine *C. jejuni* infection models in which the intestinal microbiota was modified or even depleted (Bereswill et al., 2011a; Haag et al., 2012a, b; Heimesaat et al., 2013a). It is well known that the immune system of isolator-raised germfree mice is extremely compromised and dwarfish (reviewed by Yi and Li, 2012). In order to avoid and circumvent this limitation, we eradicated the intestinal microbiota in adult mice with a maturated and fully developed immune system. This was achieved by treatment of conventionally raised mice with broad-spectrum antibiotic compounds for approximately eight weeks. As expected, *C. jejuni* colonized these gnotobiotic animals at high loads along the entire GI tract and induced pro-inflammatory immune responses in the colon (Bereswill et al., 2011a). These basic reproductions confirmed again that colonization resistance of conventional mice against *C. jejuni* is caused by the murine intestinal microbiota. Furthermore, distinct bacterial populations of mice from our animal facility are proven extraordinarily effective in eradicating not only *C. jejuni* but also apathogenic *E. coli* reference strains (Kupz et al., 2013). Most importantly colonization resistance against *E. coli* was abrogated in gnotobiotic mice generated by antibiotic treatment and in conventional mice displaying acute intestinal inflammation such as acute ileitis (Heimesaat et al., 2013c). However, peroral transplantation of the microbiota derived from the inflamed ileal lumen alone could not abrogate colonization resistance (Heimesaat et al., 2013a). The respective mouse models were feasible to investigate the essential role of metal homeostasis in the intestinal colonization capacity and pro-inflammatory potential of *E. coli* reference strains including the probiotic *E. coli* strain Nissle 1917 (Kupz et al., 2013; Bereswill et al., 2013).

Campylobacter infection in mice with human microbiota

The earlier findings that conventional mice from only certain animal facilities and mice with a defined limited microbiota (Chang and Miller, 2006) were susceptible to *C. jejuni* infection (Mansfield et al., 2007; Bell et al., 2009; MacKichan et al., 2004) indicated that not the microbiota per se but the distinct microbiota composition is essential for maintaining colonization resistance against *C. jejuni* (reviewed by Masanta et al., 2013). In order to further investigate if the host-specific intestinal microbiota composition is required for colonization resistance, we investigated *C. jejuni* infection in gnotobiotic mice (see above), which were permanently re-associated either with a complex intestinal microbiota derived from humans or from mice (the latter as a control). The human microbiota in those – with respect to their intestinal microbiota – "humanized" animals proved to be stable over several months (Bereswill et al., 2011a). Following peroral

infection *C. jejuni* stably colonized the GI tract of mice with human microbiota whereas mice re-associated with their original murine microbiota cleared the pathogen efficiently within one day after infection (Bereswill et al., 2011a). Strikingly, in mice harbouring human microbiota *C. jejuni* induced inflammatory responses mimicking key features of human campylobacteriosis.

Microbiota changes as risk factors for campylobacteriosis

Additional experiments were performed to identify risk factors for alterations in the microbiota composition, which in turn cause a breakdown of colonization resistance against *C. jejuni*. Corresponding results revealed that colonization resistance against *C. jejuni* was severely impaired in ob/ob mice with intrinsic obesity, in wildtype mice fed a hypercaloric "western" or "cafeteria" diet, in mice with acute intestinal inflammation and in conventional infant mice when infected immediately after weaning (Haag et al., 2012c; Bereswill et al., 2011b; Otto et al., 2012). Interestingly, all these conditions have elevated intestinal concentrations of enterobacteria in common. Enterobacteria and *Escherichia coli* in particular are known to accumulate at sites of intestinal inflammation, aggravate immunopathology and translocate to submucosal tissues via microulcerations (Bereswill et al., 2013). Most notably, feeding conventional mice live *E. coli* via the drinking water abrogated colonization resistance - thus rendering mice susceptible to *C. jejuni* infection (Haag et al., 2012a). This approach is easy to achieve in any murine facility and can therefore be used to study the complex interactions of *C. jejuni* and related species with *E. coli* and other gut bacteria.

Infant mice models for *Campylobacter* research

Infant mice infected with *C. jejuni* immediately after weaning have been used for decades as effective infection models for the study of campylobacteriosis. The *C. jejuni* flagellum was confirmed to serve as a key pathogenicity factor in infant mice (Diker et al., 1992) and infant mice were successfully used for the development of vaccination strategies against *C. jejuni* (Dolby and Newell, 1986; Abimiku et al., 1989). We could recently confirm that conventional infant mice are well suited for the analysis of *C. jejuni* virulence, pathogenicity and colonization capacity (Haag et al., 2012c). These animals were even suited to study long-term infection of otherwise asymptomatic *C. jejuni* carriers. The animals displayed long-term shedding of *C. jejuni* and developed extraintestinal chronic sequelae (Heimesaat et al., 2013b). Furthermore, infant mice proved useful to analyse the pathogenic potential of *C. jejuni* mutants lacking virulence genes such as the extracellular protease HtrA (Heimesaat et al., 2014a). Again, the microbiota composition of conventional infant mice was characterized by higher loads of commensal enterobacterial species such as *E. coli* as compared to three months old adult control animals.

Taken together, the "humanized" animals (with respect to the microbiota composition), mice colonized with defined bacterial species as well as gnotobiotic mice and infant mice are suited to study host responses in campylobacteriosis and to identify bacterial species or metabolic conditions involved in colonization resistance against *C. jejuni*. The important question whether gnotobiotic mice generated by antibiotic treatment can circumvent the experimental shortcomings and disadvantages of the "dwarfish" immune system in isolator-raised mice awaits further investigation. However, the knowledge of bacterial species that are protective against *C. jejuni* colonization will support a better understanding of the infectious process per se. The discovery of the underlying metabolic conditions could pave the way for novel strategies to optimize both protection or treatment of infection. Notably, the "humanized" mouse model proved useful for the study of microbiota changes, translocation of intestinal bacterial commensal species to extra-intestinal compartments and changes of distinct immune cell populations in the intestinal tract post mortem (Heimesaat et al., 2012d).

The role of innate immune receptors in acute campylobacteriosis

Toll-like receptors (TLRs) comprise essential first line components of signalling pathways involved in innate and adaptive host responses to pathogenic infections. *C. jejuni* lipo-oligosaccharide (LOS) was shown to aggravate human campylobacteriosis via TLR-4 signalling in clinical studies (Mortensen et al., 2009). This important finding further underlines that infection studies in gnotobiotic TLR-4 deficient mice (with diminished colonization resistance) are well suited to unravel the immunopathology of human campylobacteriosis in more detail. However, detailed in vivo studies on the interplay of TLRs with *C. jejuni* in the intestinal tract are scarce. In gnotobiotic mice generated by antibiotic treatment we were able to show that detection of *C. jejuni* LOS and CpG-DNA by host TLR-4 and TLR-9, respectively, was essential for mediating pro-inflammatory immune responses in the colon upon *C. jejuni* infection (Bereswill et al., 2011a). Thus gnotobiotic mice lacking defined TLRs proved useful to unravel the role of innate responses to infections with *C. jejuni*. However, classical macroscopic symptoms of human campylobacteriosis such as bloody diarrhea were lacking in those gnotobotic wildtype mice.

Acute campylobacteriosis developing in germfree IL-10-deficient mice

Rodents are known to be about thousand times more resistant against lipo-polysaccharide (LPS), LOS and other TLR-4 agonists as compared to humans (Warren et al., 2010). Thus we performed *C. jejuni* infection experiments in IL-10 deficient mice, which are known to be much more sensitive to LPS and LOS as compared to wildtype animals (Haag et al., 2012b). Due to the key role of LOS in inducing *C. jejuni* pathogenesis, the IL-10 deficient animals developed non-self limiting acute ulcerative enterocolitis with wasting symptoms and bloody diarrhea within one week following infection with *C. jejuni*, but not a commensal *E. coli* strain. Hence, we have now a *C. jejuni* induced infection model mimicking key features of campylobacteriosis in immune compromised humans. The essential role of pathogenic LOS in *C. jejuni* induced immune pathogenesis was further confirmed in

C. jejuni infected gnotobiotic IL-10 deficient mice lacking TLR-4 (Haag et al., 2012b). Overall, IL-10 deficient mice were shown highly susceptible to enteric infection and proved to be well suited to study the impact of novel virulence factors in intestinal colonization, translocation, and immunopathology. For instance, these animals were successfully used to analyse the pathogenic potential of *C. jejuni* mutants lacking the extracellular protease HtrA or the outer membrane adhesin Cj0268 (Heimesaat et al., 2014b,c; Tareen et al., 2013).

A future view of murine models for the study of *C. jejuni* induced pathology

In summary, the plethora of recent studies demonstrates that distinct murine infection models (with modified microbiota and/or immune system) are excellently suited for investigating various aspects of campylobacteriosis, including virulence traits, microbiota changes and host responses induced by *C. jejuni* and related species. We further conclude that the murine microbiota is essential for colonization resistance against *C. jejuni* and possibly other related species. Gnotobiotic mice will provide useful tools for the investigation of infection and immune pathogenesis induced by *Campylobacter* spp. Conventional mice with elevated intestinal *E. coli* loads, infant mice, mice with a human microbiota and gnotobiotic IL-10 deficient mice will allow for the study of specific features of the murine microbiota in modulating the susceptibility to *C. jejuni* and related organisms including *Arcobacter* species.

Most importantly these novel findings have promoted a renaissance of murine investigations in colonization resistance and genetic variation of *C. jejuni* (Wilson et al., 2010; Kim et al., 2012; Thomas et al., 2014). Most recently modified mice models were successfully used to study functions of the innate immune receptor nucleotide-oligonucleotide-domain 2 in campylobacteriosis (Sun and Jobin, 2014), to investigate the role of amino acid metabolism in intestinal colonization (Hofreuter et al., 2012) and to further unravel the molecular mechanisms of *C. jejuni* induced enteritis and the associated autoimmune diseases including GBS (Bell et al., 2009; Lippert et al., 2009; Sun et al., 2012; Malik et al., 2014). These novel findings confirmed the important role of LOS as a major virulence factor of *C. jejuni* as shown in mice and men. Furthermore, the analysis of T-cell responses demonstrated that *C. jejuni* induces specific Th17 mediated T-cell responses in mice (Malik et al., 2014). Because similar adaptive immune responses were also seen earlier in ex vivo infected biopsies from humans (Edwards et al., 2010), this further underlines that modified mouse infection models mimick the majority of innate and adaptive immune responses in human campylobacteriosis and thus represent a realistic and feasible perspective for *C. jejuni* research in the future.

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References

Abimiku AG, Dolby JM, Borriello SP (1989): Comparison of different vaccines and induced immune response against *Campylobacter jejuni* colonization in the infant mouse. Epidemiol Infect 102: 271–280.

Alter T, Bereswill S, Glünder G, Haag LM, Hänel I, Heimesaat MM, Lugert R, Rautenschlein S, Weber RM, Zautner AE, Gross U (2011): Campylobacteriosis of man: livestock as reservoir for *Campylobacter* species. Bundesgesundheitsblatt 54: 728–734.

Bell JA, St Charles JL, Murphy AJ, Rathinam VA, Plovanich-Jones AE, Stanley EL, Wolf JE, Gettings JR, Whittam TS, Mansfield LS (2009): Multiple factors interact to produce responses resembling spectrum of human disease in *Campylobacter jejuni* infected C57BL/6 IL-10-/- mice. BMC Microbiol. 18: 241–257.

Bereswill S, Fischer A, Dunay IR, Kühl AA, Göbel UB, Liesenfeld O, Heimesaat MM (2013): Pro-inflammatory potential of *Escherichia coli* strains K12 and Nissle 1917 in a murine model of acute ileitis. Eur J Microbiol Immunol (Bp) 3: 126–134.

Bereswill S, Fischer A, Plickert R, Haag LM, Otto B, Kühl AA, Dasti JI, Zautner AE, Muñoz M, Loddenkemper C, Gross U, Göbel UB, Heimesaat MM (2011a): Novel murine infection models provide deep insights into the "ménage à trois" of *Campylobacter jejuni*, microbiota and host innate immunity. PLoS One 6: e20953.

Bereswill S, Plickert R, Fischer A, Kühl AA, Loddenkemper C, Batra A, Siegmund B, Göbel UB, Heimesaat MM (2011b): What you eat is what you get: novel *Campylobacter* models in the quadrangle relationship between nutrition, obesity, microbiota and susceptibility to infection. Eur J Microbiol Immunol (Bp) 1: 237–248.

Bereswill S, Kist M (2003): Recent developments in *Campylobacter* pathogenesis. Curr Opin Infect Dis 16: 487–491.

Bereswill S, Kist M (2002): Molecular microbiology and pathogenesis of Helicobacter and *Campylobacter* updated: a meeting report of the 11th conference on *Campylobacter, Helicobacter* and related organisms. Mol Microbiol 45: 255–262.

Chang C, Miller JF (2006): *Campylobacter jejuni* colonization of mice with limited enteric flora. Infect Immun 74: 5261–5271.

Diker KS, Hascelik G, Diker S (1992): Colonization of infant mice with flagellar variants of *Campylobacter jejuni*. Acta Microbiol Hung 39: 133–136.

Dolby JM, Newell DG (1986): The protection of infant mice from colonization with *Campylobacter jejuni* by vaccination of the dams. J Hyg (Lond): 96:143–151.

Dorrell N, Wren BW (2007): The second century of *Campylobacter* research: recent advances, new opportunities and old problems. Curr Opin Infect Dis 20: 514–518.

Edwards LA, Nistala K, Mills DC, Stephenson HN, Zilbauer M, Wren BW, Dorrell N, Lindley KJ, Wedderburn LR, Bajaj-Elliott M (2010): Delineation of the innate and adaptive T-cell immune outcome in the human host in response to *Campylobacter jejuni* infection. PLoS One 5: e15398.

Fox JG, Rogers AB, Whary MT, Ge Z, Taylor NS, Horwitz BH, Erdman SE (2004): Gastroenteritis in NF-kappaB-deficient mice is produced with wild-type *Campylobacter jejuni* but not with *C. jejuni* lacking cytolethal distending toxin despite persistent colonization with both strains. Infect Immun 72: 1116–1125.

Haag LM, Fischer A, Otto B, Plickert R, Kühl AA, Göbel UB, Bereswill S, Heimesaat MM (2012a): Intestinal microbiota shifts towards elevated commensal *Escherichia coli* loads abrogate colonization resistance against *Campylobacter jejuni* in mice. PLoS One 7: e35988.

Haag LM, Fischer A, Otto B, Plickert R, Kühl AA, Göbel UB, Bereswill S, Heimesaat MM (2012b): *Campylobacter jejuni* induces acute enterocolitis in gnotobiotic IL-10-/- mice via Toll-like-receptor-2 and -4 signaling. PLoS One 7: e40761.

Haag LM, Fischer A, Otto B, Grundmann U, Kühl AA, Göbel UB, Bereswill S, Heimesaat MM (2012c): *Campylobacter jejuni* infection of infant mice: acute enterocolitis is followed by asymptomatic intestinal and extra-intestinal immune responses. Eur J Microbiol Immunol (Bp) 2: 2–11.

Heimesaat MM, Boelke S, Fischer A, Haag LM, Loddenkemper C, Kühl AA, Göbel UB, Bereswill S (2012d): Comprehensive postmortem analyses of intestinal microbiota changes and bacterial translocation in human flora associated mice. PLoS One 7: e40758.

Heimesaat MM, Plickert R, Fischer A, Göbel UB, Bereswill S (2013a): Can microbiota transplantation abrogate murine colonization resistance against *Campylobacter jejuni*? Eur J Microbiol Immunol (Bp) 3: 36–43.

Heimesaat MM, Haag LM, Fischer A, Otto B, Kühl AA, Göbel UB, Bereswill S (2013b): Survey of extra-intestinal immune responses in asymptomatic long-term *Campylobacter jejuni*-infected mice. Eur J Microbiol Immunol (Bp) 3: 174–182.

Heimesaat MM, Kupz A, Fischer A, Nies DH, Grass G, Göbel UB, Bereswill S (2013c): Colonization resistance against genetically modified *Escherichia coli* K12 (W3110) strains is abrogated following broad-spectrum antibiotic treatment and acute ileitis. Eur J Microbiol Immunol (Bp) 3: 222–228.

Heimesaat MM, Fischer A, Alutis M, Grundmann U, Boehm M, Tegtmeyer N, Göbel UB, Kühl AA, Bereswill S, Backert S (2014a): The impact of serine protease HtrA in apoptosis, intestinal immune responses and extra-intestinal histopathology during *Campylobacter jejuni* infection of infant mice. Gut Pathog 6: 16. doi: 10.1186/1757-4749-6-16.

Heimesaat MM, Lugert R, Fischer A, Alutis M, Kühl AA, Zautner AE, Tareen AM, Göbel UB, Bereswill S (2014b): Impact of *Campylobacter jejuni* cj0268c knockout mutation on intestinal colonization, translocation, and induction of immunopathology in gnotobiotic IL-10 deficient mice. PLoS One 9: e90148.

Heimesaat MM, Alutis M, Grundmann U, Fischer A, Tegtmeyer N, Böhm M, Kühl AA, Göbel UB, Backert S, Bereswill S (2014c): The role of serine protease HtrA in acute ulcerative enterocolitis and extra-intestinal immune responses during *Campylobacter jejuni* infection of gnotobiotic IL-10 deficient mice. Front Cell Infect Microbiol 4(77): 1–14.

Hodgson AE, McBride BW, Hudson MJ, Hall G, Leach SA (1998): Experimental campylobacter infection and diarrhoea in immunodeficient mice. J Med Microbiol 47: 799–809.

Hofreuter D, Mohr J, Wensel O, Rademacher S, Schreiber K, Schomburg D, Gao B, Galán JE (2012): Contribution of amino acid catabolism to the tissue specific persistence of *Campylobacter jejuni* in a murine colonization model. PLoS One 7: e50699.

Jesudason MV, Hentges DJ, Pongpech P (1989): Colonization of mice by *Campylobacter jejuni*. Infect Immun 57: 2279–2282.

Kim JS, Artymovich KA, Hall DF, Smith EJ, Fulton R, Bell J, Dybas L, Mansfield LS, Tempelman R, Wilson DL, Linz JE (2012): Passage of *Campylobacter jejuni* through the chicken reservoir or mice promotes phase variation in contingency genes Cj0045 and Cj0170 that strongly associates with colonization and disease in a mouse model. Microbiology 158: 1304–1316.

Kist M, Bereswill S (2001): *Campylobacter jejuni*. Contrib Microbiol 8: 150–165.

Kupz A, Fischer A, Nies DH, Grass G, Göbel UB, Bereswill S, Heimesaat MM (2013): Impact of metal ion homeostasis of genetically modified *Escherichia coli* Nissle 1917 and K12 (W3110) strains on colonization properties in the murine intestinal tract. Eur J Microbiol Immunol (Bp) 3: 229–235.

Lippert E, Karrasch T, Sun X, Allard B, Herfarth HH, Threadgill D, Jobin C (2009): Gnotobiotic IL-10; NF-kappaB mice develop rapid and severe colitis following *Campylobacter jejuni* infection. PLoS One. 4:e7413.

MacKichan JK, Gaynor EC, Chang C, Cawthraw S, Newell DG, Miller JF, Falkow S (2004): The *Campylobacter jejuni* dccRS two-component system is required for optimal in vivo colonization but is dispensable for in vitro growth. Mol Microbiol 54: 1269–1286.

Malik A, Sharma D, St Charles J, Dybas LA, Mansfield LS (2014): Contrasting immune responses mediate *Campylobacter jejuni*-induced colitis and autoimmunity. Mucosal Immunol 7: 802–817.

Mansfield LS, Bell JA, Wilson DL, Murphy AJ, Elsheikha HM, Rathinam VA, Fierro BR, Linz JE, Young VB (2007): C57BL/6 and congenic interleukin-10-deficient mice can serve as models of *Campylobacter jejuni* colonization and enteritis. Infect Immun 75: 1099–1115.

Masanta WO, Heimesaat MM, Bereswill S, Tareen AM, Lugert R, Groß U, Zautner AE (2013): Modification of intestinal microbiota and its consequences for innate immune response in the pathogenesis of campylobacteriosis. Clin Dev Immunol (doi: 10.1155/2013/526860).

Mortensen NP, Kuijf ML, Ang CW, Schiellerup P, Krogfelt KA, Jacobs BC, van Belkum A, Endtz HP, Bergman MP (2009): Sialylation of *Campylobacter jejuni* lipo-oligosaccharides is associated with severe gastro-enteritis and reactive arthritis. Microbes Infect 11: 988–994.

Otto B, Haag LM, Fischer A, Plickert R, Kühl AA, Göbel UB, Heimesaat MM, Bereswill S (2012): *Campylobacter jejuni* induces extra-intestinal immune responses via toll-like-receptor-4 signaling in conventional IL-10 deficient mice with chronic colitis. Eur J Microbiol Immunol (Bp) 2: 210–219.

Sun X, Threadgill D, Jobin C (2012): *Campylobacter jejuni* induces colitis through activation of mammalian target of rapamycin signaling. Gastroenterology 142:86–95.

Sun X, Jobin C (2014): Nucleotide-binding oligomerization domain-containing protein 2 controls host response to *Campylobacter jejuni* in Il10-/- Mice. J Infect Dis Apr 10 [Epub ahead of print].

Tareen AM, Lüder CG, Zautner AE, Groß U, Heimesaat MM, Bereswill S, Lugert R (2013): The *Campylobacter jejuni* Cj0268c protein is required for adhesion and invasion in vitro. PLoS One 8: e81069.

Thomas DK, Lone AG, Selinger LB, Taboada EN, Uwiera RR, Abbott DW, Inglis GD (2014): Comparative variation within the genome of *Campylobacter jejuni* NCTC 11168 in human and murine hosts. PLoS One 9: e88229.

Warren HS, Fitting C, Hoff E, Adib-Conquy M, Beasley-Topliffe L, Tesini B, Liang X, Valentine C, Hellman J, Hayden D, Cavaillon JM (2010): Resilience to bacterial infection: difference between species could be due to proteins in serum. J Infect Dis 201: 223–232.

Wilson DL, Rathinam VA, Qi W, Wick LM, Landgraf J, Bell JA, Plovanich-Jones A, Parrish J, Finley RL, Mansfield LS, Linz JE (2010): Genetic diversity in *Campylobacter jejuni* is associated with differential colonization of broiler chickens and C57BL/6J IL10-deficient mice. Microbiology 156: 2046–2057.

Yi P, Li L (2012): The germfree murine animal – an important animal model for research on the relationship between gut microbiota and the host. Vet Microbiol 157: 1–7.

Yrios JW, Balish E (1986a): Immune response of athymic and euthymic germfree mice to *Campylobacter* spp. Infect Immun 54: 339–346.

Yrios JW, Balish E (1986b): Colonization and infection of athymic and euthymic germfree mice by *Campylobacter jejuni* and *Campylobacter fetus* subsp. *fetus*. Infect Immun 53: 378–383.

Yrios JW, Balish E (1986c): Pathogenesis of *Campylobacter* spp. in athymic and euthymic germfree mice. Infect Immun 53: 384–392.

Young VB, Schauer DB, Fox JG (2000): Animal models of *Campylobacter* infection. In: Nachamkin I, BlaserMJ, (editors): *Campylobacter*, 2nd ed. Washington, DC: ASM Press, 287–302.

Youssef M, Corthier G, Goossens H, Tancrede C, Henry-Amar M, Andremont A (1987): Comparative translocation of enteropathogenic *Campylobacter* spp. and *Escherichia coli* from the intestinal tract of gnotobiotic mice. Infect Immun 55: 1019–1021.

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