Virulence

ISSN: 2150-5594 (Print) 2150-5608 (Online) Journal homepage: https://www.tandfonline.com/loi/kvir20

New tricks new ways: Exploitation of a multifunctional enzyme arginase by pathogens

Amit Lahiri, Priyanka Das & Dipshikha Chakravortty

To cite this article: Amit Lahiri, Priyanka Das & Dipshikha Chakravortty (2010) New tricks new ways: Exploitation of a multifunctional enzyme arginase by pathogens , Virulence, 1:6, 563-565, DOI: 10.4161/viru.1.6.13623

To link to this article: https://doi.org/10.4161/viru.1.6.13623

Virulence 1:6, 563-565; November/December 2010; © 2010 Landes Bioscience

New tricks new ways

Exploitation of a multifunctional enzyme arginase by pathogens

Amit Lahiri, Priyanka Das and Dipshikha Chakravortty*

Center for Infectious Disease Research and Biosafety Laboratories; Department of Microbiology and Cell Biology; Indian Institute of Science; Bangalore, Karnataka India

Introduction

Arginases are highly conserved family of enzymes across the kingdom.[1] Discovered by Kossler and Dahler in 1904, arginase is no longer a mere enzyme responsible only for urea cycle but is described to be involved in various other ways. Interest in arginase was stimulated by research that demonstrated its role in nitric oxide (NO) synthesis. Modulation of arginase by pathogenic organisms now seems to be a hot evasion strategy employed by various pathogens. To be precise, arginase can be called as the key enzyme, which modulates the immune system and thus pathogenesis. Though the knowledge of mammalian arginine metabolism and its role in immunology is vast, the critical evasion strategy by pathogenic organisms by using the host arginase enzyme is still a mystery.

There are two isoforms of vertebrate arginase, both of which catalyze the conversion of L-arginine to L-ornithine and urea but which differ with regard to tissue distribution and subcellular localization. Type I arginase (arginase I), a cytosolic enzyme, is highly expressed in liver as a component of the urea cycle, whereas type II arginase (arginase II) is a mitochondrial enzyme that is expressed to varying degrees in many cell types.[1,2] Furthermore, various pathogens also code for their own arginase which might as well behave like its host counterpart. In this article, we will summarize the recent findings that deal with both host and pathogenic arginase, mainly focusing on the role in modulating the virulence of any pathogen.

Arginase Mediated Modulatation of Pathogenesis at Various Levels

Arginine is the common substrate for both inducible nitric oxide synthase (iNOS) and arginase. Thus, if a pathogen channelizes the host arginine toward arginase pathway, there might be a reduced substrate for iNOS, thereby generating reduced amount of nitric oxide (NO). If the NO radicals are not generated, then the pathogen can have a very secure niche in the infected host. *Salmonella typhimurium*,[3] *Mycobacteria tuberculosis*,[4] *Leishmania mexicana*[5] and *Schistosoma mansoni*[6] use this strategy to survive in the host. Salmonella upregulates arginase II in the macrophages. Blocking arginase increases the substrate L-arginine availability to iNOS for production of more nitric oxide and perhaps peroxynitrite molecules in the infected cells allowing better killing of virulent Salmonella in a NO-dependent manner. Inhibition of arginase activity in mice by NOHA during the course of Salmonella infection reduces the bacterial burden and delays the disease outcome in a NO-dependent manner. Lower *M. tuberculosis* load was observed in the Arg-1-deficient mice. In the same report it was observed that the liver granuloma from BCG-infected mice produced greater bactericidal nitrotyrosine when host arginase was knocked out.[4]

Interestingly, pathogens like *Helicobacter pylori (H. pylori*) possess a gene *rocF,* which encodes for arginase and again quenches away the l-arginine pool from iNOS, thereby generating less amount of NO. Production of arginase by *H. pylori* is further pivotal for its virulence as the *rocF* mutant is non-pathogenic.[7] Further, high level of host arginase was obtained from the *H. pylori*-infected gastric tissue. These data clearly indicate the importance of both bacterial and host arginase in case of Helicobacter infection.[8] Arginase can also directly act on the T cells and reduce the host-mediated killing. *H. pylori* arginase can impair the T cell function by reducing the CD3ζ expression during infection.[9]

Polyamines are also produced as one of the byproduct in the arginase pathway which can be used as a nutrient by few pathogens. Leishmania encode their own arginase which has considerable potential to modulate infectivity and disease pathogenesis. For example, Leishmania uses polyamines for its growth[10] and utilizes the parasite coded arginase for this purpose. The arginase knockout parasites retained the ability to differentiate normally to the infective metacyclic stage, and were able to induce progressive disease following inoculation into susceptible BALB/c mice but less efficiently than the WT parasites. In an interesting report, the infectivity and impact on host cellular immune response in vitro and in vivo of wild-type *L. major* with that of arginase knockout *L. major*. It was observed that arginase knockout *L. major* are impaired in their macrophage infectivity in vitro independent of host inducible NO synthase activities. The arginase knock outs were impaired in vivo infection, resulting in delayed onset of lesion development, attenuated pathology and low parasite burden.[11] Specific inhibition of the host arginase I by nor-NOHA treatment decreases the parasite load and delays

*Correspondence to: Dipshikha Chakravortty; Email: dipa@mcbl.iisc.ernet.in
Submitted: 07/14/10; Revised: 09/13/10; Accepted: 09/13/10
Previously published online: www.landesbioscience.com/journals/virulence/article/13623
DOI: 10.4161/viru.1.6.13623

lesion development in the susceptible BALB/c mice. On the other hand in the resistant C57BL/6 mice ornithine supplementation increases the susceptibility of infection, clearly suggesting the fact that in cutaneous leishmaniasis host arginase pathway is hijacked by the parasite for polyamine acquisition.[12] These host- and parasite-derived polyamine can also modulate the production of pro-inflammatory cytokines in the infected host.[13] *H. pylori* further utilize the polyamine spermine to restrain immune response in activated macrophages.[14]

Arginase is also responsible for collagen synthesis and tissue regeneration.[15] Hence, it is possible that the pathogens associated with cancer like *H. pylori* and Hepatitis C virus (HCV) may use arginase to modulate the cell cycle thereby leading to cancerous state. In fact, siRNA against arginase inhibited the ability of HCV to promote hepatocellular growth.[16] In case of periodontal diseases, heightened level of salivary arginase was observed which came down after antibiotic treatment.[17] This shows that arginase perhaps can be used as a diagnostic marker for several infections.

The most important question is does pathogen modulate the arginase in the dendritic cells (DC). DC being an efficient antigen presenting cell, is targeted by successful pathogen. The modulation of arginase level in DC by the pathogens can perhaps lead to inhibition of antigen presentation of impaired T cell function. In many infectious diseases where the immunity is impaired, could be due to arginase. *Staphylococcus aureus* is a pathogen carrying its own arginase and might modulate host arginase as well. Looking at the havoc the *Staphylococcus aureus* causes, it would be worth to look into the role of arginase in pathogenesis.

Future Direction

The constant tussle between the host and the pathogen to regulate the arginase isoforms determine the outcome of several infectious diseases. Therapeutic intervention aimed at arginase may prove beneficial in many infectious diseases. Especially, in case of *Mycobacterium tuberculosis*, arginase plays a pivotal role. Further in case of co-infection with HIV, inhibition of arginase can restore the cell-mediated immunity thereby helping the host. It has been reported that age plays an important role in arginase-mediated L-arginine metabolism. In younger individuals, increased induction of arginase may result in a more permissive environment for growth of parasite like Leishmania, thereby leading to increased severity of the disease.[18] Hence, targeting arginase might be a promising therapeutic intervention against leishmaniasis and other infectious diseases, in children and young adults. However, as arginase is a multi-functional enzyme with two isoforms, care should be taken to restore its normal function during the therapy. Wide variety of bacterial, viral and parasitic pathogens uses arginase as an evasion strategy. Many pathogens possess their own arginase which functions similar to the mammalian arginase. Hence, this gives us further direction to develop new therapeutic drugs of molecules which can inhibit the function of arginase, thereby restoring the antimicrobial activity of the cells.

One more point to look into is how the pathogenic arginases that are intracellular and not secreted outside in the cytosol can get access to the host arginine pool. It is well documented that Schistosoma arginase is localized to the head of the organism and is not secreted upon infection[19] or the *Helicobacter pylori* arginase which is again intracellular.[20] In this regard, a study related to Candida pathogenesis should be taken into account. In order to escape from macrophages after being ingested, Candida employs a very fascinating strategy of inducing its own intracellular arginase and urea amidolyase to achieve hyphal switching. Candida induces two other endogenous arginase as well that are secreted out. These extracellular arginases may provide survival benefit to Candida by reducing nitrosative stress via quenching the iNOS substrate arginine.[21] Studies should focus whether similar secretory arginases are present in other pathogens as well.

Note of Caution

Arginase is an essential enzyme by which the host is protected from the detrimental effect of NO and gets polyamines for its cellular functions.[22] Ammonia, which is very toxic for the cells, gets cleared by Urea cycle. T lymphocyte functions are regulated by arginases and arginine. Hence, on one hand where arginase is physiologically an important enzyme, on the other the same is used by the pathogen. To make the situation more complicated it is known that the arginase knockout mice are embryonic lethal. If arginase inhibitors are used as a therapeutic, it may lead to ammonia toxicity or abundance NO generation. Developing bacterial specific arginase inhibitors can be fruitful.

References

1. Wu G, Morris SM Jr. Arginine metabolism: Nitric oxide and beyond. Biochem J 1998; 336:1-17.
2. Morris SM Jr. Enzymes of arginine metabolism. J Nutr 2004; 134:2743-7.
3. Lahiri A, Das P, Chakravortty D. Arginase modulates Salmonella induced nitric oxide production in RAW264.7 macrophages and is required for Salmonella pathogenesis in mice model of infection. Microbes Infect 2008; 10:1166-74.
4. El Kasmi KC, Qualls JE, Pesce JT, Smith AM, Thompson RW, Henao-Tamayo M, et al. Toll-like receptor-induced arginase 1 in macrophages thwarts effective immunity against intracellular pathogens. Nat Immunol 2008; 9:1399-406.
5. Gaur U, Roberts SC, Dalvi RP, Corraliza I, Ullman B, Wilson ME. An effect of parasite-encoded arginase on the outcome of murine cutaneous leishmaniasis. J Immunol 2007; 179:8446-53.
6. Thompson RW, Pesce JT, Ramalingam T, Wilson MS, White S, Cheever AW, et al. Cationic amino acid transporter-2 regulates immunity by modulating arginase activity. PLoS Pathog 2008; 4:1000023.
7. Gobert AP, McGee DJ, Akhtar M, Mendz GL, Newton JC, Cheng Y, et al. *Helicobacter pylori* arginase inhibits nitric oxide production by eukaryotic cells: A strategy for bacterial survival. Proc Natl Acad Sci USA 2001; 98:13844-9.
8. McGee DJ, Radcliff FJ, Mendz GL, Ferrero RL, Mobley HL. *Helicobacter pylori* rocF is required for arginase activity and acid protection in vitro but is not essential for colonization of mice or for urease activity. J Bacteriol 1999; 181:7314-22.
9. Zea AH, Rodriguez PC, Culotta KS, Hernandez CP, DeSalvo J, Ochoa JB, et al. L-Arginine modulates CD3zeta expression and T cell function in activated human T lymphocytes. Cell Immunol 2004; 232:21-31.
10. Reguera RM, Balana-Fouce R, Showalter M, Hickerson S, Beverley SM. *Leishmania major* lacking arginase (ARG) are auxotrophic for polyamines but retain infectivity to susceptible BALB/c mice. Mol Biochem Parasitol 2009; 165:48-56.
11. Muleme HM, Reguera RM, Berard A, Azinwi R, Jia P, Okwor IB, et al. Infection with arginase-deficient *Leishmania major* reveals a parasite number-dependent and cytokine-independent regulation of host cellular arginase activity and disease pathogenesis. J Immunol 2009; 183:8068-76.
12. Iniesta V, Carcelen J, Molano I, Peixoto PM, Redondo E, Parra P, et al. Arginase I induction during *Leishmania major* infection mediates the development of disease. Infect Immun 2005; 73:6085-90.
13. Munder M, Eichmann K, Moran JM, Centeno F, Soler G, Modolell M. Th1/Th2-regulated expression of arginase isoforms in murine macrophages and dendritic cells. J Immunol 1999; 163:3771-7.

14. Bussiere FI, Chaturvedi R, Cheng Y, Gobert AP, Asim M, Blumberg DR, et al. Spermine causes loss of innate immune response to *Helicobacter pylori* by inhibition of inducible nitric-oxide synthase translation. J Biol Chem 2005; 280:2409-12.
15. Shearer JD, Richards JR, Mills CD, Caldwell MD. Differential regulation of macrophage arginine metabolism: a proposed role in wound healing. Am J Physiol 1997; 272:181-90.
16. Cao W, Sun B, Feitelson MA, Wu T, Tur-Kaspa R, Fan Q. Hepatitis C virus targets overexpression of arginase I in hepatocarcinogenesis. Int J Cancer 2009; 124:2886-92.
17. Gheren LW, Cortelli JR, Rodrigues E, Holzhausen M, Saad WA. Periodontal therapy reduces arginase activity in saliva of patients with chronic periodontitis. Clin Oral Investig 2008; 12:67-72.
18. Muller I, Hailu A, Choi BS, Abebe T, Fuentes JM, Munder M, et al. Age-related alteration of arginase activity impacts on severity of leishmaniasis. PLoS Negl Trop Dis 2008; 2:235.
19. Fitzpatrick JM, Fuentes JM, Chalmers IW, Wynn TA, Modolell M, Hoffmann KF, et al. *Schistosoma mansoni* arginase shares functional similarities with human orthologs but depends upon disulphide bridges for enzymatic activity. Int J Parasitol 2009; 39:267-79.
20. Yoshiyama H, Nakazawa T. Unique mechanism of *Helicobacter pylori* for colonizing the gastric mucus. Microbes Infect 2000; 2:55-60.
21. Ghosh S, Navarathna DH, Roberts DD, Cooper JT, Atkin AL, Petro TM, et al. Arginine-induced germ tube formation in *Candida albicans* is essential for escape from murine macrophage line RAW 264.7. Infect Immun 2009; 77:1596-605.
22. Das P, Lahiri A, Chakravortty D. Modulation of the arginase pathway in the context of microbial pathogenesis: a metabolic enzyme moonlighting as an immune modulator. PLoS Pathog 6:1000899.