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TOPIC HIGHLIGHT

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Interplay between intestinal alkaline phosphatase, diet, gut microbes and immunity

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Abstract

Intestinal alkaline phosphatase (IAP) plays an essential role in intestinal homeostasis and health through interactions with the resident microbiota, diet and the gut. IAP's role in the intestine is to dephosphorylate toxic microbial ligands such as lipopolysaccharides, unmethylated cytosine-guanosine dinucleotides and flagellin as well as extracellular nucleotides such as uridine diphosphate. IAP's ability to detoxify these ligands is essential in protecting the host from sepsis during acute inflammation and chronic inflammatory conditions such as inflammatory bowel disease. Also important in these complications is IAP's ability to regulate the microbial ecosystem by forming a complex relationship between microbiota, diet and the intestinal mucosal surface. Evidence reveals that diet alters IAP expression and activity and this in turn can influence the gut microbiota and homeostasis. IAP's ability to maintain a healthy gastrointestinal tract has accelerated research on its potential use as a therapeutic agent against a multitude of diseases. Exogenous IAP has been shown to have beneficial effects when administered during ulcerative colitis, coronary bypass surgery and sepsis. There are currently a handful of human clinical trials underway investigating the effects of exogenous IAP during sepsis, rheumatoid arthritis and heart surgery. In light of these findings IAP has been marked as a novel agent to help treat a variety of other inflammatory and infectious diseases. The purpose of this review is to highlight the essential characteristics of IAP in protection and maintenance of intestinal homeostasis while addressing the intricate interplay between IAP, diet, microbiota and the intestinal epithelium.

Key words: Intestinal alkaline phosphatase; Inflammation; Infection; Lipopolysaccharides; Inflammatory disease; Intestinal microbiota; Diet

Core tip: Intestinal alkaline phosphatase (IAP) is important for intestinal health. IAP's role in the intestine encompasses both protection from systemic infections and chronic inflammatory diseases such as inflammatory bowel disease. There is a complex interplay occurring between IAP, diet, microbiota and the intestinal epithelium which has accelerated research on IAP as a potential therapeutic against these inflictions. The purpose of this review is to highlight the essential characteristics of IAP in maintaining homeostasis in the intestines while addressing the complex interplay between IAP, diet, microbiota and the intestinal epithelium.

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INTESTINAL ALKALINE PHOSPHATASE

Alkaline phosphatases (AP) are hydrolase enzymes that catalyze the breakdown of monophosphate esters by removal of their phosphate groups. AP are grouped into two classes: the tissue non-specific alkaline phosphatases (TNAP), which are expressed in bone; liver; and kidney[1] and tissue specific isozymes which include placental alkaline phosphatase (PLAP), germ cell alkaline phosphatase; and intestinal alkaline phosphatase (IAP).

IAP is a homodimer with each subunit comprised of two Zn^{++} and one Mg^{++} ions[2], which are essential in maintaining the structural integrity and catalytic activity of the enzyme. IAP functions predominantly in alkaline conditions with optimal activity occurring at a pH of 9.7[3]. IAP is ubiquitously expressed by enterocytes in the duodenum[1] and thereafter to a much lesser extent in the jejunum, ileum and colon[4,5] and is largely absent in the stomach[5,6]. The TNAP isoform is also normally found at low levels in the healthy colon[7]. IAP molecules exist in high concentrations within lumenal vesicles secreted by enterocytes on the brush border of the microvilli (Figure 1). Though largely membrane bound, small levels of IAP are also released bidirectionally into the blood as well as the lumen[8] where they may travel throughout the intestinal tract. Interestingly, IAP levels in humans differ across blood types with types O and B showing the highest levels of IAP while type A shows the lowest[9]. Infiltrating neutrophils have been shown to contain AP granules[10], which are thought to be of the TNAP isoform[11]. Recently however, using an antibody specific to the intestinal isoform we found infiltrating immune cells in mouse colons to be localized with IAP during during infectious colitis[12].

IAP is the most evolutionarily conserved of the APs sharing 89.5% of its amino acid sequences with PLAP[13], in humans these genes are located on chromosome 2[14]. In contrast, TNAP types share only 50% of their amino acids with IAP[13] and are located on chromosome 4[14]. This indicates an evolutionary split between tissue specific and the tissue non-specific isotypes. The IAP enzyme in humans is encoded by the *ALPI* gene[15] while two isozymes have been identified in mice: duodenal IAP (dIAP) and global IAP (gIAP). Mouse dIAP and gIAP share 79% genetic homology[16] and are encoded by the *Akp*3 and *Akp*6 genes, respectively[17,18]. There are also two known IAP isozymes in rats: IAP I and IAP Ⅱ encoded by the *alpi*1 and *alpi*2 genes, respectively. Phylogenetic comparisons show that mouse gIAP is similar to both mouse dIAP and rat IAP I but not to human IAP. In general, most murine studies have focused on IAP generally and not the specific isoforms and as a result the specific function of each isozyme is poorly understood. Recent reviews have highlighted the importance of IAP in gut homeostasis[19,20].

IAP, LIPOPOLYSACCHARIDE AND INFECTION

Lipopolysaccharide (LPS) is a major component of the cell wall of Gram-negative bacteria. Its aberrant presence in the blood (endotoxemia) can stimulate strong inflammatory responses as seen during infection-induced sepsis but at lower levels can also stimulate low levels of chronic inflammatory responses like those seen during obesity, diabetes and heart disease. Circulating LPS binds to toll-like receptor (TLR) 4 on immunocytes activating the secretion of pro-inflammatory cytokines *via* the master regulatory nuclear factor-kappaB (NF-κB) pathway[21]. During infection, if the pathogen can overwhelm the host defenses, increasing cytokine levels can ultimately lead to septic shock and death. IAP plays a crucial role in preventing sepsis against enteric infection by dephosphorylating the endotoxin harboring lipid A moiety of LPS[22]. IAP is a mucosal defense factor that limits bacterial translocation across the mucosal barrier into the mesenteric lymph nodes[7,21]. It has been shown that administration of bovine IAP prevents translocation of bacteria in mice infected with *Salmonella enterica* serovar Typhimurium[23]. We recently demonstrated the association of IAP activity and survival during enteric infection in a model of high-fat feeding. We compared various high-fat diets (HFD) and found that while ω-6 polyunsaturated fatty acid (PUFA) exacerbated damage associated with colitis, increased IAP activity was associated with protection of the mice against sepsis during *Citrobacter rodentium* infection. In contrast, isocaloric and isonitrogenous diets supplemented with ω-3 PUFA led to impaired IAP activity associated with an increased sepsis-associated mortality during infection[12]. Beyond IAP's local activity at the gut mucosa, about 1%-2% of IAP is released into the bloodstream or the gastrointestinal lumen, which may broaden its range of action against both local and systemic infections[23]. In support of this, IAP has been shown to be active in feces indicating that IAP released into the intestinal lumen retains its function as it moves through the gut[21].

IAP AND INFLAMMATION

In addition to LPS, IAP can also dephosphorylate other potentially pro-inflammatory ligands such as adenosine triphosphate (ATP)[24], uridine diphosphate (UDP) nucleotides released from damaged cells[25,26], unmethylated cytosine-guanosine dinucleotides (CpG); a component of bacterial DNA[27] and flagellin; a protein in bacterial flagellum[28]. These ligands or microbe-associated molecular patterns (MAMPs) activate TLR 2, 3, 5 and 9 on immune and epithelial cells[29] inducing downstream inflammatory responses. To mitigate these responses, IAP expression increases through activation of the inflammatory mediator Resolvin E1 (RvE1)[30]. RvE1 is a lipid mediator derived from ω-3 PUFAs eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)[31]. RvE1 acts as an agonist of the G protein-coupled receptor Chemerin receptor

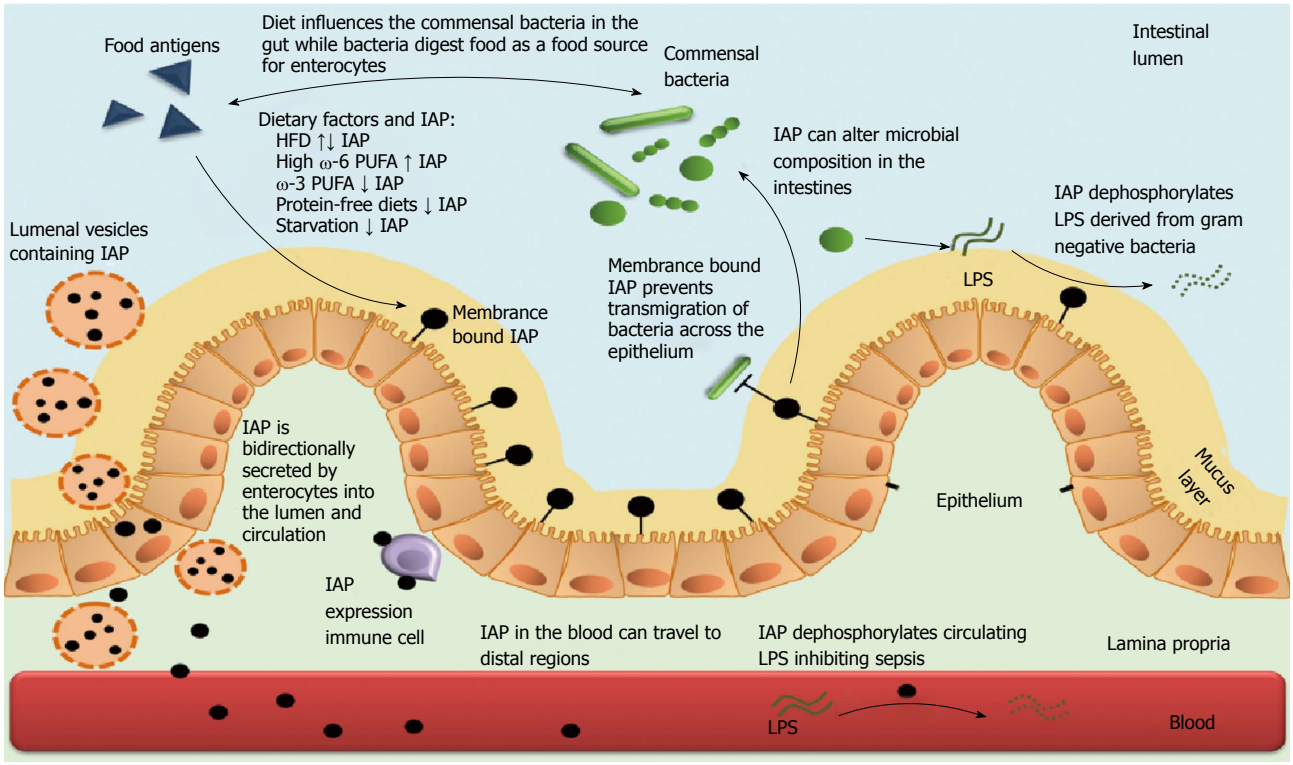

Figure 1 Intestinal alkaline phosphatase regulates gut homeostasis. Intestinal alkaline phosphatase (IAP) secreted by the enterocytes plays a vital role in various physiological functions in and around the intestine. Though mainly membrane-bound, IAP can be found both in the lumen and blood. High concentration of IAP molecules are present in protein-rich lumenal vesicles on the lumenal and apical side of the epithelium. IAP dephosphorylates both lumenal and circulating lipopolysaccharides (LPS) derived from the cell wall of gram negative bacteria effectively eliminating their toxic constituent. Preliminary work from our lab and others have shown IAP expressed on infiltrated immunocytes in the lamina propria. IAP is also crucial in preventing the transmigration of bacteria across the epithelium layer, preventing downstream activation of immunocytes and the subsequent inflammatory responses. Through its complex relationship with food, commensal bacteria and immune cells, IAP plays an essential role in gut homeostasis.

23 (Chem R23) found on neutrophils, macrophages and dendritic cells; which in turn can attenuate immune cell transmigration. Chem R23 receptors are also present on intestinal epithelial cells, where their activation increases epithelial IAP expression and activity[30,32]. In a series of studies Campbell *et al*[30,32] showed IAP mRNA expression to be reduced during DSS-induced colitis in mice while the addition of RvE1 was able to induce IAP mRNA expression, resulting in significantly attenuated disease severity. They further showed the addition of an IAP inhibitor to reverse the RvE1-induced protection against DSS, highlighting the role of RvE1 in mediating IAP expression and activity. This would suggest that EPA-derived RvE1, as seen in EPA-rich diets, can activate IAP and therefore protect against colitis. In contrast, we found that fish oil, rich in EPA and DHA, supplemented on a background diet high in ω-6 PUFA resulted in lower IAP activity associated with sepsis during an infectious model of colitis[12]. These studies highlight the complex role that diet has on the function of IAP in the gut or perhaps the various and specific roles of IAP during colonic damage *vs* enteric infection. Inflammation itself can regulate *IAP* gene expression and activity, though the mechanisms are not yet fully understood. Malo *et al*[33] showed that the pro-inflammatory cytokines interleukin (IL)-1β and tumor necrosis factor (TNF)-α inhibit *ALPI* gene expression in HT-29 human colon adenocarcinoma cells. This suggests that cytokine-mediated gene silencing may have important implications in altering epithelial health and function during intestinal inflammation. More research is required to understand the relationship between IAP and its involvement during acute and chronic inflammation.

IAP AND CHRONIC INFLAMMATORY DISEASES

Altered IAP expression has been implicated in many chronic inflammatory states such as IBD, celiac disease and obesity. IBD patients have reduced *ALPI* gene expression in inflamed tissues[4]. Oral administration of IAP during murine colitis reduces colonic inflammation as demonstrated by decreases in mRNA levels of the key pro-inflammatory cytokine TNF-α and nitric oxide producing inducible nitric oxide synthase (iNOS)[5]. This suggests that exogenous IAP may be protective against colitis. It has been reported that patients with ulcerative colitis have reduced colonic pH levels[34]. One report using chicken IAP showed that the enzyme is inactivated at pH 6; however the addition of Zn ions resulted in reactivation of IAP even at pH 4.5. This indicates that under certain conditions IAP can function at even low pH levels[35], such as those seen during colitis. Another factor

important in IBD is increased levels of extracellular nucleotides such as ATP and UDP which has been directly associated with intestinal inflammation[25]. These nucleotides are released during cellular damage and death[26] such as seen during active IBD, and bind to receptors on macrophages, epithelial cells and infiltrating T-cells. By means of dephosphorylation, IAP blocks the subsequent inflammatory responses induced by these nucleotides[24,25].

Gut homeostasis is vital for maintaining a balanced metabolism, not surprisingly then endogenous IAP has been recently implicated in metabolic syndromes[36]. In an elegant series of studies by Kaliannan *et al*[36], it was shown that IAP knock-out (KO) mice display features of metabolic syndrome such as: obesity, elevated blood glucose, endotoxemia, glucose intolerance and hyperinsulinemia. Supplementation with oral IAP was able to prevent and reverse these conditions in both the KO mice as well as in models of HFD-induced metabolic syndrome.

IAP, MICROBIOTA AND INTESTINAL HOMEOSTASIS

It has been suggested that IAP can directly regulate the ecology of the gut microbiota. Oral administration of IAP in mice showed a reduction in severity rates of *Salmonella* Typhimurium and *Clostridium difficile* (*C. difficile*) infections[37]. These changes were associated with IAP's ability to rapidly restore the commensal gut microbiota. A plausible explanation is through IAP's capacity to control pH, which in turn alters bacterial growth. IAP achieves this by dephosphorylating luminal phosphates such as ATP[24,37]. This results in increased levels of bicarbonate ion secretion which is important for maintaining homeostasis and gut barrier function[6]. It should be noted however, that *Salmonella* Typhimurium and *C. difficile* colonization in the gut in these models required prior antibiotic treatments in order to deplete the microbiota, potentially obscuring IAP's true role in reducing inflammatory responses[38]. In another study, Bates *et al*[38] showed that IAP reduced inappropriate inflammatory responses in germ-free zebrafish larvae compared to control larvae incubated with a stereospecific IAP inhibitor, L-phenylalanine. IAP-rich larvae had decreased neutrophils in their gut epithelium compared to the IAP inhibited larvae which were able to recruit neutrophils in response to LPS stimulation. In a series of experiments, Malo *et al*[39] showed IAP-KO mice to display an altered ecosystem with reduced total fecal bacteria compared to their wild type (WT) counterparts. In their experiments, WT mice had higher levels of Lactobacillaceae group and *Escherichia coli* (*E. coli*) expression compared to KO mice as the absence of IAP prevented *E. coli* from colonizing the gut. Further, oral administration of IAP resulted in increased colonization of *E. coli* in the IAP-KO mice. This is important because the loss of commensal bacteria such as *E. coli* can create a vulnerable gut environment allowing for colonization of potential pathobionts[40]. A follow-up study by the same group confirmed that ATP and other nucleotide triphosphates inhibit the growth of commensal bacterial and that introduction of IAP can promote their growth[24]. These studies confirm the interaction between IAP and the microbiota as a novel dimension in which IAP regulates intestinal health.

INTERPLAY BETWEEN FOOD AND IAP

Diet has been regarded as an important factor in many pathological conditions such as IBD, metabolic syndrome, diabetes and infection. IAP is regulated by dietary macronutrients as well as fasting. Mice fasted for two days showed significant decreases in IAP expression and as a result decreased LPS-dephosphorylating activity compared to non-fasted mice[21]. This is important in the context of trophic enteral feeding because as suggested by Goldberg *et al*[21] starvation in critically ill patients may lead to IAP downregulation and therefore an increased susceptibility to pathogenic infection. The re-introduction of food in these mice resulted in the recovery of IAP expression, suggesting an adaptive response of IAP to food availability. IAP is important during HF feeding as it is involved in the rate-limiting step of fatty acids transport across the plasma membrane into the enterocytes[18,41]. IAP secretion increases in response to HFD[16], likely in a negative feedback manner to negate the resulting increases in intestinal LPS levels[36,42]. Another study showed that gIAP, but not dIAP, protein expression significantly increased in response to an HFD with 45% fat content [77% from saturated fatty acids (SFA)][17]. HFD increases the uptake of fatty acids by enterocytes through a gIAP-dependent mechanism. In contrast, another study using an HFD consisting of 36.3% SFA caused reduced IAP activity and increased TLR 4 activation in diet-induced obese prone rats[42]. Therefore, increased consumption of HFD induces an inflammatory response and results in downregulation of IAP expression. The differences between these two studies suggest that the amount of fat in a diet can discordantly influence IAP regulation. Further studies are required to investigate the specific effects of SFA on IAP. Work from our lab has shown that mice fed diets high in ω-6 PUFA (20% w/w) have increased IAP expression within the lamina propria. In attempt to mitigate colitic responses, ω-3 PUFA was supplemented in these HFD which resulted in decreased IAP expression and activity[12]. However, these mice unexpectedly suffered from higher incidences of sepsis-associated mortality. Taken together, these findings suggests that in addition to the amount of fat, the consumption of particular dietary fatty acids can also alter IAP expression and activity, a vital consequence to the host during infection. The effects of HFD on IAP expression may also be potentially linked to one particular IAP isozyme. dIAP knock out (*Akp3*$^{-/-}$) mice showed increased weight gain during a HFD diet compared to WT[18]. These phenotypic responses however, may be compensatory actions of intact gIAP (*Akp6*) in these mice. Therefore, AKP6-KO experiments are needed to provide more insight, as each

Table 1 Summary of completed and ongoing clinical human trials using exogenous bovine intestinal alkaline phosphatase

Area of study	Study design	Route of administration	Study phase	Treatment effect	Sample size/ Estimated enrollment[1]	Ref.[1]
Completed trials						
Coronary artery bypass	Randomized, placebo	Intravenous	2	Increased endogenous alkaline phosphatase release	32	Kats *et al*[47] (2012)
Severe sepsis and septic shock on acute kidney injury	Double-blind, randomized and placebo	Intravenous	2	Improved renal function	36	Heemskerk *et al*[49] (2009)
Moderate to severe ulcerative colitis	Uncontrolled	Oral	2	Short term improvement of moderate/severe ulcerative colitis	20	Lukas *et al*[50] (2010)
Sepsis in patients with acute kidney injury (AKI)	Double-blind, randomized and placebo	Intravenous	2	Renal protection from sepsis in patients with AKI	36	Pickers *et al*[48] (2012)
Ongoing trials						
Acute rheumatoid arthritis	Non-randomized	Subcutaneous	1 and 2	Ongoing study	10	NCT01416493[2]
Safety and efficacy during heart surgery	Randomized double-blind, placebo-controlled	Intravenous	3	Ongoing study	228	NCT01144611[2]

[1]Refers to ongoing trials; [2]The trial numbers associated with ongoing clinical studies was obtained from clinicaltrials.gov.

isozyme may function differently during inflammation and infection.

Protein consumption may also play an important role in regulating IAP. Rats fed protein-free diets showed a 36%-38% reduction in IAP activity[43]. In this experiment however, the rat diets were high in starch and therefore IAP levels may potentially be altered due to high levels of carbohydrates rather than from the lack of protein. These results taken together suggest that there is a complex interplay between diet and IAP activity, and that customized diets may be beneficial in regulating IAP levels.

IAP AS A THERAPEUTIC AGENT

There is growing evidence that IAP administration during infection and inflammation may be a novel therapeutic strategy in reducing disease complications. IAP supplementation may also be an effective tool in battling the rising problem of antibiotic related infections such as *C. difficile* associated diseases (CDAD). In support of this, oral administration of IAP concurrently with antibiotics in mice was shown to completely prevent CDAD as well as other enteric pathogens like *Salmonella* Typhimurium[37]. This is critical because administering antibiotics can induce a microbial imbalance in the gut, known as dysbiosis. This creates a favorable environment for pathogens and increases host susceptibility to infection.

Another therapeutic application of IAP for decreasing pro-inflammatory responses is in neonates suffering from necrotizing enterocolitis (NEC)[44]. Administration of IAP in rats reduced serum levels of pro-inflammatory cytokines TNF-α, IL-5 and IL-1β during NEC. These levels resemble that of the healthy controls indicating that exogenous IAP has the ability to downregulate the immune response and normalize inflammation in these infants, however this did not decrease the overall incidence of NEC. Tuin *et al*[5] showed that supplementation with IAP during rat colitis also significantly reduced pro-inflammatory markers TNF-α, IL-1β and iNOS. As a result there was significant improvement in colonic morphology, though no improvement in clinical symptoms such as: weight loss, diarrhea or rectal bleeding. IAP was also shown to be beneficial in mice with cystic fibrosis (CF); a known chronic pulmonary disease associated with intestinal bacterial overgrowth[45]. Exogenous IAP restored intestinal permeability and reduced 80% of intestinal bacteria overgrowth in mice with CF[46]. Most clinical trials to date report administration of IAP through intravenous injections as to avoid major degradation and digestion in the stomach and upper intestinal tract. A method that has not been reported in humans is rectal enemas as this allows direct IAP delivery to the large intestine. Also to date, exogenous IAP in the form of enteric coated or delayed-release capsules has not been reported. IAP appears to have extensive therapeutic potential and has relatively safe pharmacokinetic properties, as a result several clinical human trials have begun. There are currently four completed IAP human trials[47-50] and two ongoing studies which includes research on acute rheumatoid arthritis and safety and efficacy of IAP use during heart surgery (Table 1).

CONCLUSION

IAP's role in catalyzing the breakdown of monophosphate esters is well established, however, only within the last decade has the role of IAP as a key regulator of inflammation, infection and gut microbiota been explored. IAP also plays a key role in protecting the host during chronic inflammatory diseases. IAP expression and activ-

ity is altered as a result of its complex interplay between dietary factors, the microbiota and the host. In light of these findings IAP has been marked as a potential therapeutic agent to help treat a variety of inflammatory and infectious diseases.

REFERENCES

1 **Narisawa S**, Huang L, Iwasaki A, Hasegawa H, Alpers DH, Millán JL. Accelerated fat absorption in intestinal alkaline phosphatase knockout mice. *Mol Cell Biol* 2003; **23**: 7525-7530 [PMID: 14560000]

2 **Muginova SV**, Zhavoronkova AM, Polyakov AE, Shekhovtsova TN. Application of alkaline phosphatases from different sources in pharmaceutical and clinical analysis for the determination of their cofactors; zinc and magnesium ions. *Anal Sci* 2007; **23**: 357-363 [PMID: 17372382]

3 **Metwalli OM**, Mourand FE. Studies on organ-specific alkaline phosphatases in relation to their diagnostic value. *Z Ernahrungswiss* 1980; **19**: 154-158 [PMID: 7445572]

4 **Molnár K**, Vannay A, Szebeni B, Bánki NF, Sziksz E, Cseh A, Győrffy H, Lakatos PL, Papp M, Arató A, Veres G. Intestinal alkaline phosphatase in the colonic mucosa of children with inflammatory bowel disease. *World J Gastroenterol* 2012; **18**: 3254-3259 [PMID: 22783049 DOI: 10.3748/wjg.v18.i25.3254]

5 **Tuin A**, Poelstra K, de Jager-Krikken A, Bok L, Raaben W, Velders MP, Dijkstra G. Role of alkaline phosphatase in colitis in man and rats. *Gut* 2009; **58**: 379-387 [PMID: 18852260 DOI: 10.1136/gut.2007.128868]

6 **Akiba Y**, Mizumori M, Guth PH, Engel E, Kaunitz JD. Duodenal brush border intestinal alkaline phosphatase activity affects bicarbonate secretion in rats. *Am J Physiol Gastrointest Liver Physiol* 2007; **293**: G1223-G1233 [PMID: 17916646 DOI: 10.1152/ajpgi.00313.2007]

7 **Martínez-Moya P**, Ortega-González M, González R, Anzola A, Ocón B, Hernández-Chirlaque C, López-Posadas R, Suárez MD, Zarzuelo A, Martínez-Augustin O, Sánchez de Medina F. Exogenous alkaline phosphatase treatment complements endogenous enzyme protection in colonic inflammation and reduces bacterial translocation in rats. *Pharmacol Res* 2012; **66**: 144-153 [PMID: 22569414 DOI: 10.1016/j.phrs.2012.04.006]

8 **McConnell RE**, Higginbotham JN, Shifrin DA, Tabb DL, Coffey RJ, Tyska MJ. The enterocyte microvillus is a vesicle-generating organelle. *J Cell Biol* 2009; **185**: 1285-1298 [PMID: 19564407 DOI: 10.1083/jcb.200902147]

9 **Bayer PM**, Hotschek H, Knoth E. Intestinal alkaline phosphatase and the ABO blood group system--a new aspect. *Clin Chim Acta* 1980; **108**: 81-87 [PMID: 7449139]

10 **Detmers PA**, Zhou D, Powell D, Lichenstein H, Kelley M, Pironkova R. Endotoxin receptors (CD14) are found with CD16 (Fc gamma RIII) in an intracellular compartment of neutrophils that contains alkaline phosphatase. *J Immunol* 1995; **155**: 2085-2095 [PMID: 7543538]

11 **Smith GP**, Harris H, Peters TJ. Studies of the biochemical and immunological properties of human neutrophil alkaline phosphatase with comparison to the established alkaline phosphatase isoenzymes. *Clin Chim Acta* 1984; **142**: 221-230 [PMID: 6499206]

12 **Ghosh S**, DeCoffe D, Brown K, Rajendiran E, Estaki M, Dai C, Yip A, Gibson DL. Fish oil attenuates omega-6 polyunsaturated fatty acid-induced dysbiosis and infectious colitis but impairs LPS dephosphorylation activity causing sepsis. *PLoS One* 2013; **8**: e55468 [PMID: 23405155 DOI: 10.1371/journal.pone.0055468]

13 **Henthorn PS**, Raducha M, Edwards YH, Weiss MJ, Slaughter C, Lafferty MA, Harris H. Nucleotide and amino acid sequences of human intestinal alkaline phosphatase: close homology to placental alkaline phosphatase. *Proc Natl Acad Sci USA* 1987; **84**: 1234-1238 [PMID: 3469665]

14 **Millán JL**. Alkaline phosphatase as a reporter of cancerous transformation. *Clin Chim Acta* 1992; **209**: 123-129 [PMID: 1395034]

15 **Buchet R**, Millán JL, Magne D. Multisystemic functions of alkaline phosphatases. *Methods Mol Biol* 2013; **1053**: 27-51 [PMID: 23860646 DOI: 10.1007/978-1-62703-562-0_3]

16 **Alpers DH**, Zhang Y, Ahnen DJ. Synthesis and parallel secretion of rat intestinal alkaline phosphatase and a surfactant-like particle protein. *Am J Physiol* 1995; **268**: E1205-E1214 [PMID: 7611397]

17 **Lynes M**, Narisawa S, Millán JL, Widmaier EP. Interactions between CD36 and global intestinal alkaline phosphatase in mouse small intestine and effects of high-fat diet. *Am J Physiol Regul Integr Comp Physiol* 2011; **301**: R1738-R1747 [PMID: 21900644 DOI: 10.1152/ajpregu.00235.2011]

18 **Narisawa S**, Hoylaerts MF, Doctor KS, Fukuda MN, Alpers DH, Millán JL. A novel phosphatase upregulated in Akp3 knockout mice. *Am J Physiol Gastrointest Liver Physiol* 2007; **293**: G1068-G1077 [PMID: 17901166 DOI: 10.1152/ajpgi.00073.2007]

19 **Lallès JP**. Intestinal alkaline phosphatase: novel functions and protective effects. *Nutr Rev* 2014; **72**: 82-94 [PMID: 24506153 DOI: 10.1111/nure.12082]

20 **Lallès JP**. Intestinal alkaline phosphatase: multiple biological roles in maintenance of intestinal homeostasis and modulation by diet. *Nutr Rev* 2010; **68**: 323-332 [PMID: 20536777 DOI: 10.1111/j.1753-4887.2010.00292.x]

21 **Goldberg RF**, Austen WG, Zhang X, Munene G, Mostafa G, Biswas S, McCormack M, Eberlin KR, Nguyen JT, Tatlidede HS, Warren HS, Narisawa S, Millán JL, Hodin RA. Intestinal alkaline phosphatase is a gut mucosal defense factor maintained by enteral nutrition. *Proc Natl Acad Sci USA* 2008; **105**: 3551-3556 [PMID: 18292227 DOI: 10.1073/pnas.0712140105]

22 **Beumer C**, Wulferink M, Raaben W, Fiechter D, Brands R, Seinen W. Calf intestinal alkaline phosphatase, a novel therapeutic drug for lipopolysaccharide (LPS)-mediated diseases, attenuates LPS toxicity in mice and piglets. *J Pharmacol Exp Ther* 2003; **307**: 737-744 [PMID: 12970380 DOI: 10.1124/jpet.103.056606]

23 **Chen KT**, Malo MS, Beasley-Topliffe LK, Poelstra K, Millan JL, Mostafa G, Alam SN, Ramasamy S, Warren HS, Hohmann EL, Hodin RA. A role for intestinal alkaline phosphatase in the maintenance of local gut immunity. *Dig Dis Sci* 2011; **56**: 1020-1027 [PMID: 20844955 DOI: 10.1007/s10620-010-1396-x]

24 **Malo MS**, Moaven O, Muhammad N, Biswas B, Alam SN, Economopoulos KP, Gul SS, Hamarneh SR, Malo NS, Teshager A, Mohamed MM, Tao Q, Narisawa S, Millán JL, Hohmann EL, Warren HS, Robson SC, Hodin RA. Intestinal alkaline phosphatase promotes gut bacterial growth by reducing the concentration of luminal nucleotide triphosphates. *Am J Physiol Gastrointest Liver Physiol* 2014; **306**: G826-G838 [PMID: 24722905 DOI: 10.1152/ajpgi.00357.2013]

25 **Moss AK**, Hamarneh SR, Mohamed MM, Ramasamy S, Yammine H, Patel P, Kaliannan K, Alam SN, Muhammad N, Moaven O, Teshager A, Malo NS, Narisawa S, Millán JL, Warren HS, Hohmann E, Malo MS, Hodin RA. Intestinal alkaline phosphatase inhibits the proinflammatory nucleotide uridine diphosphate. *Am J Physiol Gastrointest Liver Physiol* 2013; **304**: G597-G604 [PMID: 23306083 DOI: 10.1152/ajpgi.00455.2012]

26 **Di Virgilio F**, Chiozzi P, Ferrari D, Falzoni S, Sanz JM, Morelli A, Torboli M, Bolognesi G, Baricordi OR. Nucleotide receptors: an emerging family of regulatory molecules in blood cells. *Blood* 2001; **97**: 587-600 [PMID: 11157473]

27 **Hemmi H**, Takeuchi O, Kawai T, Kaisho T, Sato S, Sanjo H, Matsumoto M, Hoshino K, Wagner H, Takeda K, Akira S. A Toll-like receptor recognizes bacterial DNA. *Nature* 2000;

408: 740-745 [PMID: 11130078 DOI: 10.1038/35047123]
28 **Chen KT**, Malo MS, Moss AK, Zeller S, Johnson P, Ebrahimi F, Mostafa G, Alam SN, Ramasamy S, Warren HS, Hohmann EL, Hodin RA. Identification of specific targets for the gut mucosal defense factor intestinal alkaline phosphatase. *Am J Physiol Gastrointest Liver Physiol* 2010; **299**: G467-G475 [PMID: 20489044 DOI: 10.1152/ajpgi.00364.2009]
29 **Cario E**. Bacterial interactions with cells of the intestinal mucosa: Toll-like receptors and NOD2. *Gut* 2005; **54**: 1182-1193 [PMID: 15840688 DOI: 10.1136/gut.2004.062794]
30 **Campbell EL**, Louis NA, Tomassetti SE, Canny GO, Arita M, Serhan CN, Colgan SP. Resolvin E1 promotes mucosal surface clearance of neutrophils: a new paradigm for inflammatory resolution. *FASEB J* 2007; **21**: 3162-3170 [PMID: 17496159 DOI: 10.1096/fj.07-8473com]
31 **El Kebir D**, Gjorstrup P, Filep JG. Resolvin E1 promotes phagocytosis-induced neutrophil apoptosis and accelerates resolution of pulmonary inflammation. *Proc Natl Acad Sci USA* 2012; **109**: 14983-14988 [PMID: 22927428 DOI: 10.1073/pnas.1206641109]
32 **Campbell EL**, MacManus CF, Kominsky DJ, Keely S, Glover LE, Bowers BE, Scully M, Bruyninckx WJ, Colgan SP. Resolvin E1-induced intestinal alkaline phosphatase promotes resolution of inflammation through LPS detoxification. *Proc Natl Acad Sci USA* 2010; **107**: 14298-14303 [PMID: 20660763 DOI: 10.1073/pnas.0914730107]
33 **Malo MS**, Biswas S, Abedrapo MA, Yeh L, Chen A, Hodin RA. The pro-inflammatory cytokines, IL-1beta and TNF-alpha, inhibit intestinal alkaline phosphatase gene expression. *DNA Cell Biol* 2006; **25**: 684-695 [PMID: 17233117]
34 **Fallingborg J**, Christensen LA, Jacobsen BA, Rasmussen SN. Very low intraluminal colonic pH in patients with active ulcerative colitis. *Dig Dis Sci* 1993; **38**: 1989-1993 [PMID: 8223071]
35 **Kunitz M**. Chicken intestinal alkaline phosphatase. I. The kinetics and thermodynamics of reversible inactivation. 2. Reactivation by zinc ions. *J Gen Physiol* 1960; **43**: 1149-1169 [PMID: 14412759]
36 **Kaliannan K**, Hamarneh SR, Economopoulos KP, Nasrin Alam S, Moaven O, Patel P, Malo NS, Ray M, Abtahi SM, Muhammad N, Raychowdhury A, Teshager A, Mohamed MM, Moss AK, Ahmed R, Hakimian S, Narisawa S, Millán JL, Hohmann E, Warren HS, Bhan AK, Malo MS, Hodin RA. Intestinal alkaline phosphatase prevents metabolic syndrome in mice. *Proc Natl Acad Sci USA* 2013; **110**: 7003-7008 [PMID: 23569246 DOI: 10.1073/pnas.1220180110]
37 **Alam SN**, Yammine H, Moaven O, Ahmed R, Moss AK, Biswas B, Muhammad N, Biswas R, Raychowdhury A, Kaliannan K, Ghosh S, Ray M, Hamarneh SR, Barua S, Malo NS, Bhan AK, Malo MS, Hodin RA. Intestinal alkaline phosphatase prevents antibiotic-induced susceptibility to enteric pathogens. *Ann Surg* 2014; **259**: 715-722 [PMID: 23598380 DOI: 10.1097/SLA.0b013e31828fae14]
38 **Bates JM**, Akerlund J, Mittge E, Guillemin K. Intestinal alkaline phosphatase detoxifies lipopolysaccharide and prevents inflammation in zebrafish in response to the gut microbiota. *Cell Host Microbe* 2007; **2**: 371-382 [PMID: 18078689 DOI: 10.1016/j.chom.2007.10.010]
39 **Malo MS**, Alam SN, Mostafa G, Zeller SJ, Johnson PV, Mohammad N, Chen KT, Moss AK, Ramasamy S, Faruqui A, Hodin S, Malo PS, Ebrahimi F, Biswas B, Narisawa S, Millán JL, Warren HS, Kaplan JB, Kitts CL, Hohmann EL, Hodin RA. Intestinal alkaline phosphatase preserves the normal homeostasis of gut microbiota. *Gut* 2010; **59**: 1476-1484 [PMID: 20947883 DOI: 10.1136/gut.2010.211706]
40 **Manichanh C**, Rigottier-Gois L, Bonnaud E, Gloux K, Pelletier E, Frangeul L, Nalin R, Jarrin C, Chardon P, Marteau P, Roca J, Dore J. Reduced diversity of faecal microbiota in Crohn's disease revealed by a metagenomic approach. *Gut* 2006; **55**: 205-211 [PMID: 16188921 DOI: 10.1136/gut.2005.073817]
41 **Clark SB**, Holt PR. Rate-limiting steps in steady-state intestinal absorption of trioctanoin-1-14C. Effect of biliary and pancreatic flow diversion. *J Clin Invest* 1968; **47**: 612-623 [PMID: 5637147 DOI: 10.1172/JCI105757]
42 **de La Serre CB**, Ellis CL, Lee J, Hartman AL, Rutledge JC, Raybould HE. Propensity to high-fat diet-induced obesity in rats is associated with changes in the gut microbiota and gut inflammation. *Am J Physiol Gastrointest Liver Physiol* 2010; **299**: G440-G448 [PMID: 20508158 DOI: 10.1152/ajpgi.00098.2010]
43 **Montoya CA**, Leterme P, Lalles JP. A protein-free diet alters small intestinal architecture and digestive enzyme activities in rats. *Reprod Nutr Dev* 2006; **46**: 49-56 [PMID: 16438914 DOI: 10.1051/rnd: 2005063]
44 **Riggle KM**, Rentea RM, Welak SR, Pritchard KA, Oldham KT, Gourlay DM. Intestinal alkaline phosphatase prevents the systemic inflammatory response associated with necrotizing enterocolitis. *J Surg Res* 2013; **180**: 21-26 [PMID: 23158403 DOI: 10.1016/j.jss.2012.10.042]
45 **Norkina O**, Burnett TG, De Lisle RC. Bacterial overgrowth in the cystic fibrosis transmembrane conductance regulator null mouse small intestine. *Infect Immun* 2004; **72**: 6040-6049 [PMID: 15385508 DOI: 10.1128/IAI.72.10.6040-6049.2004]
46 **De Lisle RC**, Mueller R, Boyd M. Impaired mucosal barrier function in the small intestine of the cystic fibrosis mouse. *J Pediatr Gastroenterol Nutr* 2011; **53**: 371-379 [PMID: 21970994 DOI: 10.1097/MPG.0b013e318219c397]
47 **Kats S**, Brands R, Hamad MA, Seinen W, Scharnhorst V, Wulkan RW, Schönberger JP, Oeveren WV. Prophylactic treatment with alkaline phosphatase in cardiac surgery induces endogenous alkaline phosphatase release. *Int J Artif Organs* 2012; **35**: 144-151 [PMID: 22395920 DOI: 10.5301/ijao.5000039]
48 **Pickkers P**, Heemskerk S, Schouten J, Laterre PF, Vincent JL, Beishuizen A, Jorens PG, Spapen H, Bulitta M, Peters WH, van der Hoeven JG. Alkaline phosphatase for treatment of sepsis-induced acute kidney injury: a prospective randomized double-blind placebo-controlled trial. *Crit Care* 2012; **16**: R14 [PMID: 22269279 DOI: 10.1186/cc11159]
49 **Heemskerk S**, Masereeuw R, Moesker O, Bouw MP, van der Hoeven JG, Peters WH, Russel FG, Pickkers P. Alkaline phosphatase treatment improves renal function in severe sepsis or septic shock patients. *Crit Care Med* 2009; **37**: 417-23, e1 [PMID: 19114895 DOI: 10.1097/CCM.0b013e31819598af]
50 **Lukas M**, Drastich P, Konecny M, Gionchetti P, Urban O, Cantoni F, Bortlik M, Duricova D, Bulitta M. Exogenous alkaline phosphatase for the treatment of patients with moderate to severe ulcerative colitis. *Inflamm Bowel Dis* 2010; **16**: 1180-1186 [PMID: 19885903 DOI: 10.1002/ibd.21161]

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