



IMPORTANCE OF AUTOTAXIN IN LIVER DISORDERS AND INTERACTION WITH OTHER LIPID MEDIATORS AND POSSIBLY NATRIURETIC PEPTIDES

Manoj G. Tyagi*

Department of Biotechnology, Vellore Institute of Technology, Vellore 632004, Tamilnadu.

*Corresponding Author: Manoj G Tyagi

Department of Biotechnology, Vellore Institute of Technology, Vellore 632004, Tamilnadu.

Article Received on 23/03/2016

Article Revised on 14/04/2016

Article Accepted on 05/05/2016

ABSTRACT

Natriuretic peptides and in particular the B-type natriuretic peptide (BNP) stimulates the proliferation, differentiation, and lipolysis of preadipocytes by upregulation of the levels of expression of its receptor NPR1 and key genes enriched in the glycerolipid metabolic pathway. These lipids accumulate in the liver. Recent studies suggest that BNP may stimulate lipid metabolism by affecting various enzymes. On the other hand, Lysophosphatidic acid (LPA) is a bioactive lipid mediator involved in many biological processes. Concentrations of the major LPA species in mouse plasma decrease following administration of a potent selective inhibitor of the LPA-generating lysophospholipase D (autotaxin) identifying an active mechanism for removal of LPA from the circulation. This review examines the effect of Autotaxin in liver disorders and possible implications for the involvement of natriuretic peptides in the lipid metabolism in liver.

KEYWORDS: B-type natriuretic peptide, Liver, Lipids, Lysophosphatidic acid, Autotaxin.

INTRODUCTION

The natriuretic peptide BNP was initially discovered in the porcine brain, but the largest concentrations of BNP are found in the heart. It is a peptide with 32 amino acids, synthesised in the ventricles as a response to stretching of the myocytes and/or pressure overload. It is released as an active hormone and as an inactive N-terminal fragment (NT-pro-BNP).^[1] Upon its release in the blood flow, BNP has numerous physiological actions, their net effect being to reduce pre- and post-load. Specifically, BNP produces a decreased vascular tone by relaxing the smooth muscles, leading to a decrease in post-load. In addition, it induces a movement of fluid into the interstitial space, thus leading to a decrease in pre-load.^[2] BNP reduces the proliferation of fibroblasts and smooth muscle cells, sympathetic nervous activity, water and salt retention, release of the antidiuresis hormone, and synthesis of aldosterone and its release from the adrenal glands. There is accumulating evidence of the role of BNP in hepatic lipid metabolism.^[3] There is growing importance of the LPAs which exhibit a broad range of biological activities that are initiated by LPA selective G-protein coupled cell-surface receptors. Some LPA responses may also be mediated by the nuclear peroxisome proliferator gamma receptor and the receptor for advanced glycan end products.^[4,5] LPA is present in blood plasma and accumulates in human atheromas and experimentally induced atherosclerotic lesions in mice. Studies using LPA receptor deficient mice and LPA-directed small molecule therapeutics identify roles for

LPA signaling in athero-thrombosis and vascular injury responses. LPA is actively accumulated in the liver. Studies conducted using an isolated liver perfusion system indicated that ~90% of LPA accumulates in the liver during the first pass. The majority of liver-associated LPA, at least at early times following intravenous administration, is found in association with non-parenchymal cells of the liver which include sinusoidal epithelial cells, Kupffer cells and stellate cells.^[6] A role for the liver in elimination and metabolism of circulating LPA is supported by observations that plasma LPA levels are increased in patients with chronic hepatitis C and correlated strongly with the severity of liver dysfunction and fibrosis. LPA also plays a causative role in cholestatic pruritis characteristic of liver diseases that include biliary cirrhosis, primary sclerosing cholangitis, and intra-hepatic cholestasis of pregnancy in which post-hepatic elimination is impaired. While in one case these pathologies may be linked to increases in circulating ATX levels it is plausible that impairment of the role of the liver in elimination of circulating LPA could also contribute to increases in plasma LPA levels.^[7]

B-type natriuretic peptide and liver fat metabolism

Recent studies report that higher natriuretic peptide levels are characterized by decreased visceral and liver fat and increased lower body fat irrespective of age, gender, race, and the status of obesity. This suggested that there might be a link between the heart and adipose

tissue distribution that is mediated through the natriuretic peptides. A study of 608 Japanese patients with type 2 diabetes indicated that there was an inverse relationship between BNP and body mass index (BMI), with a more robust inverse association between BNP and visceral fat compared with other measures of adiposity.^[8] Treatments of cells with BNP led to enhanced proliferation and differentiation of cells and glycerine concentration, and mRNA expression of its receptor natriuretic peptide receptor 1 (NPR1) was upregulated significantly. In cells exposed to the BNP, 480 differentially expressed genes were identified compared with controls without BNP treatment. At least four different genes known to be related to lipid metabolism (diacylglycerol kinase; endothelial lipase; 1-acylglycerol-3-phosphate O-acyltransferase 1; and 1-acylglycerol-3-phosphate O-acyltransferase 2) were enriched in the glycerolipid metabolism pathway and expressed differentially. It was concluded from this study that, BNP stimulates the proliferation, differentiation, and lipolysis of preadipocytes through upregulation of the levels of expression of its receptor NPR1 and key genes enriched in the glycerolipid metabolic pathway. Brain-specific NPR-B deletion prevented body weight gain induced by a high-fat diet (HFD), and the mesenteric fat and liver weights were significantly decreased in BND mice fed an HFD. The decreased liver weight in BND mice was attributed to decreased lipid accumulation in the liver, which was confirmed by histologic findings and lipid content. Gene expression analysis revealed a significant decrease in the mRNA expression levels of CD36, Fsp27, and Mogat1 in the liver of BND mice, and uncoupling protein 2 mRNA expression was significantly lower in the mesenteric fat of BND mice fed an HFD than in that of control mice.^[9] This difference was not observed in the epididymal or subcutaneous fat. Furthermore, natriuretic peptides induce lipid oxidation and energy expenditure in mice and men. Increased hepatic lipid oxidation generates beta-hydroxybutyrate, which also affects food intake. Recently, it has been demonstrated that higher BNP levels correlate with favorable adiposity profiles, including reduced deposition of visceral and liver fat. The pattern of body fat distribution is an independent factor associated with metabolic syndrome in the elderly population, including dyslipidemia.^[10,11] Therefore, BNP might affect the lipid metabolism.

General procedure of Lipid extraction: Lipids can be extracted from 50-100 µl of sample (plasma or tissue homogenate) which is added to a 5 x 100 borosilicate tube containing 2 ml methanol, 1 ml chloroform, 0.45 ml 0.1M HCl. Tubes were vortexed for 5 minutes. 1 ml chloroform and 1.3 ml of 0.1M HCl were added and tubes are vortexed again for 5 min at 2500 rpm. After centrifugation the organic phase is transferred to a 4ml glass vial and evaporated under N₂. Samples were resuspended in 100ul methanol, vortexed, and stored in autosamplers at -20°C for HPLC/ESI/MS/MS analysis.

Autotaxin and liver disease

The lysophospholipase D stimulator autotaxin represents the secreted form of ectonucleotide pyrophosphatase (ENPP2) and plays a critical role in diverse physiological conditions, such as vascular and neuronal development, during pregnancy or lymphocyte migration.^[12] Autotaxin distinctly exhibits a lysophospholipase D (LPD) activity through which it hydrolyzes lysophosphatidylcholine (LPC) into lysophosphatidic acid (LPA)^[13] as shown in Fig.1. Autotaxin is widely expressed in tissues such as brain, placenta or high endothelial venules.^[14,15] In heterozygous autotaxin-null mice, both the lysoPLD activity and the LPA concentrations were about 50 % of those observed in wild-type mice, whereas complete knock-out of autotaxin is embryonic lethal due to blood vessel abnormalities, showing that autotaxin is responsible for the bulk of LPA production in blood.^[16] As ATX levels are also increased in serum of patients with other cholestatic disorders, particularly in those suffering from pruritus^[17,18], It is hypothesized that a factor capable of increasing ATX expression or reducing its clearance accumulates in cholestatic patients. Further studies are warranted to identify this factor and the source of circulating ATX levels. In a recent study by Wanvisa Udomsinprasert et al^[19] it was found that autotoxin levels were almost double than that of controls in Jaundice patients thus highlighting their importance in liver disorders.

LPA and liver disease

It is possible that the rate of elimination of intravenously administered LPA does not characterize the behaviour of the bulk of LPA in plasma some observations raise the possibility that at least a fraction of plasma LPA is rapidly being accumulated and metabolized in the liver. This may simply reflect a mechanism for scavenging albumin-bound lysophospholipids from the plasma for re-entry into hepatic pathways of lipid metabolism or it could impact on systemic or localized LPA signalling.^[20] Recent results also suggest an explanation for observations linking LPA to pathologies linked with liver dysfunction. A role for the liver in elimination and metabolism of circulating LPA is supported by observations that plasma LPA levels are increased in patients with chronic hepatitis C and correlated strongly with the severity of liver dysfunction and fibrosis. LPA also plays a causative role in cholestatic pruritis characteristics of liver diseases that include biliary cirrhosis, primary sclerosing cholangitis, and intrahepatic cholestasis of pregnancy in which post-hepatic biliary elimination is impaired. While in one case these pathologies may be linked to increases in circulating ATX levels it is plausible that impairment of the role of the liver in elimination of circulating LPA could also contribute to increases in plasma LPA levels.^[21,22]

Natriuretic peptides and lipid metabolism

Natriuretic peptides (NPs) act through natriuretic peptide receptor type A, B and C (NPRA, NPRB, NPRC) to increase protein kinase G (PKG) activity. Stimulation of

intracellular lipolysis is dependent upon PKA and PKG-mediated phosphorylation of LD-associated protein.^[23] Atrial natriuretic and B-type natriuretic peptides bind to NPR-A receptors on adipose tissue and stimulate lipolysis, promote browning of adipocytes, regulate body fat distribution by activation of peroxisome proliferator-activated receptor gamma (PPAR γ) gene expression, and enhance adiponectin secretion from adipocytes.^[24-26]

As a result, mice that overexpress or are treated with exogenous infusions of natriuretic peptide exhibit reduced fat mass, improved glucose tolerance, and enhanced energy expenditure, suggesting that the lipid mobilizing effects of natriuretic peptides may have salutary consequences of body fat metabolism and distribution.^[27]

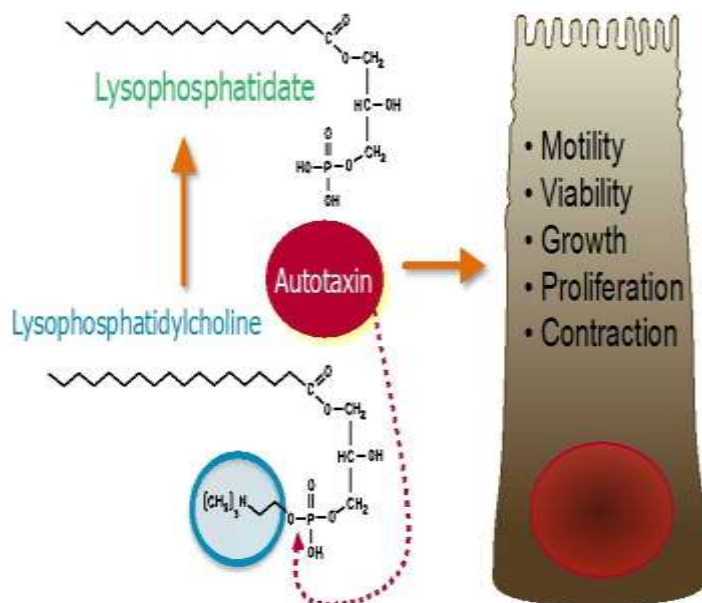


Fig.1 Courtesy: D N. Brindley, M G.K. Benesch and M M. Murph. "Melanoma - Current Clinical Management and Future Therapeutics", book edited by Mandi Murph, ISBN 978-953-51-2036-0,chapter 12, 2015

CONCLUSIONS

It is now clear that the Autotaxin contributes effectively in liver disorders and is a possible biological marker. It is also possibly interacting with the natriuretic peptides. It appears from recent findings that the association between natriuretic peptides and adiposity due to visceral lipid metabolism could be bidirectional. Experimental and observational results suggest that increased body mass leads to decreased natriuretic peptide levels and, conversely, that weight loss increases natriuretic peptide levels. The development of abdominal obesity may result in a vicious cycle, with inhibition of natriuretic peptide-mediated lipolysis and perpetuation of visceral and liver fat accumulation, ultimately causing an adverse adiposity profile and metabolic disease. An interaction of the autotoxin-LPA cascade with the natriuretic peptides is suggested in both the hepatic dysfunction and obesity related manifestations.

ACKNOWLEDGMENTS

I am thankful to Dr. Andreas E Kremer, Department of Medicine, Friedrich-Alexander-University of Erlangen-Nuremberg, Erlangen, Germany, for his advise about the theme of this research article.

REFERENCES

1. Schlueter N, de Sterke A, Willmes DM, Spranger J, Jordan J, Birkenfeld AL. Metabolic actions of natriuretic peptides and therapeutic potential in the

metabolic syndrome. *Pharmacol Ther*, 2014; 144: 12–27.

2. Neeland IJ, Winders BR, Ayers CR, Das SR, Chang AY, Berry JD, Amit Khera, Darren K. McGuire, Gloria L. Vega, James A. de Lemos, Aslan T. Turer. Higher Natriuretic Peptide Levels Associate With a Favorable Adipose Tissue Distribution Profile. *J Am Coll Cardiol*, 2013; 62: 752–60.
3. Y Yamashita, N Yamada-Goto, G Katsuura, YOchi, Y Kanai, Y Miyazaki, K Kuwahara, N Kanamoto, M Miura, A Yasoda, K Ohinata, N Inagaki, K Nakao. Brain-specific natriuretic peptide receptor-B deletion attenuates high-fat diet-induced visceral and hepatic lipid deposition in mice. 2016, *Peptides* (In Press)
4. Morris, A. J., and S. S. Smyth. Lysophosphatidic acid and cardiovascular disease: seeing is believing. *J. Lipid Res.*, 2013; 54: 1153-1155.
5. Morris, A. J., S. Selim, A. Salous, and S. S. Smyth. Blood relatives: dynamic regulation of bioactive lysophosphatidic acid and sphingosine-1-phosphate metabolism in the circulation. *Trends Cardiovasc Med.*, 2009; 19: 135-140.
6. Haseruck, N., W. Erl, D. Pandey, G. Tigyi, P. Ohlmann, C. Ravanat, C. Gachet, and W. Siess. The plaque lipid lysophosphatidic acid stimulates platelet activation and platelet-monocyte aggregate formation

- in whole blood: involvement of P2Y1 and P2Y12 receptors. *Blood*, 2004; 103: 2585-2592.
7. Panchatcharam, M., S. Miriyala, F. Yang, M. Rojas, C. End, C. Vallant, A. Dong, K. Lynch, J.Chun, A. J. Morris, and S. S. Smyth. Lysophosphatidic acid receptors 1 and 2 play roles in regulation of vascular injury responses but not blood pressure. *Circ Res.*, 2008; 103: 662-670.
 8. Sugisawa T, Kishimoto I, Kokubo Y, Nagumo A, Makino H, Miyamoto Y, et al. Visceral fat is negatively associated with B-type natriuretic peptide levels in patients with advanced type 2 diabetes. *Diabetes Res Clin Pract*, 2010; 89: 174–80.
 9. Miyashita K, Itoh H, Tsujimoto H, et al. Natriuretic peptides/cGMP/cGMP-dependent protein kinase cascades promote muscle mitochondrial bio-genesis and prevent obesity. *Diabetes*, 2009; 58: 2880-2892
 10. Bordicchia M, Liu D, Amri EZ, et al. Cardiac natriuretic peptides act via p38 MAPK to induce the brown fat thermogenic program in mouse and human adipocytes. *J Clin Invest*, 2012; 122: 1022
 11. Moolenaar WH, Perrakis A. Insights into autotaxin: how to produce and present a lipid mediator. *Nat Rev Mol Cell Biol*, 2011; 12: 674–679.
 12. Goodpaster BH, Krishnaswami S, Harris TB, Katsiaras A, Kritchevsky SB, Simonsick EM, et al. Obesity, regional body fat distribution, and the metabolic syndrome in older men and women. *Arch Intern Med.*, 2005; 165: 777–83
 13. Tokumura A, Majima E, Kariya Y, et al. Identification of human plasma lysophospholipase D, a lysophosphatidic acid-producing enzyme, as autotaxin, a multifunctional phosphodiesterase. *J Biol Chem*, 2002; 277: 39436–42.
 14. Iwasawa Y, Fujii T, Nagamatsu T, et al. Expression of autotaxin, an ectoenzyme that produces lysophosphatidic acid, in human placenta. *Am J Reprod Immunol*, 2009; 62: 90–5.
 15. Nakasaki T, Tanaka T, Okudaira S, et al. Involvement of the lysophosphatidic acid-generating enzyme autotaxin in lymphocyte endothelial cell interactions. *Am J Pathol*, 2008; 173: 1566–76.
 16. van Meeteren LA, Ruurs P, Stortelers C, et al. Autotaxin, a secreted lysophospholipase D, is essential for blood vessel formation during development. *Mol Cell Biol*, 2006; 26:5015–22.
 17. Kremer AE, van Dijk R, Leckie P, Schaap FG, Kuiper EM, Mettang T, et al. Serum autotaxin is increased in pruritus of cholestasis, but not of other origin, and responds to therapeutic interventions. *Hepatology*, 2012; 56: 1391–1400.
 18. Kremer, A. E., J. J. Martens, W. Kulik, F. Rueff, E. M. Kuiper, H. R. van Buuren, K. J. van Erpecum, J. Kondrackiene, J. Prieto, C. Rust, V. L. Geenes, C. Williamson, W. H. Moolenaar, U. Beuers, and R. P. Oude Elferink. Lysophosphatidic acid is a potential mediator of cholestatic pruritus. *Gastroenterology*, 2010; 139: 1008-1018.
 19. W Udomsinprasert, S Honsawek, W Anomasiri, V Chongsrisawat, P Vejchapipat, and Y Poovorawan. Serum autotaxin levels correlate with hepatic dysfunction and severity in postoperative biliary atresia. *Biomarkers*, 2015; 20(1): 89–94.
 20. Watanabe, N., H. Ikeda, K. Nakamura, R. Ohkawa, Y. Kume, J. Aoki, K. Hama, S. Okudaira, M. Tanaka, T. Tomiya, M. Yanase, K. Tejima, T. Nishikawa, M. Arai, H. Arai, M. Omata, K. Fujiwara, and Y. Yatomi. Both plasma lysophosphatidic acid and serum autotaxin levels are increased in chronic hepatitis C. *J Clin Gastroenterol*, 2007; 41: 616-623.
 21. Tigyi, G. Aiming drug discovery at lysophosphatidic acid targets. *Br. J. Pharmacol*, 2010; 161: 241-270.
 22. Ikeda H, Yatomi Y, Yanase M, et al. Effects of lysophosphatidic acid on proliferation of stellate cells and hepatocytes in culture. *Biochem Biophys Res Commun*, 1998; 248: 436–40.
 23. Sengenès C, Berlan M, De Glisezinski I, Lafontan M, Galitzky J. Natriuretic peptides: a new lipolytic pathway in human adipocytes. *FASEB J*, 2000; 14: 1345–51.
 24. Tsukamoto O, Fujita M, Kato M, et al. Natriuretic peptides enhance the production of adiponectin in human adipocytes and in patients with chronic heart failure. *J Am Coll Cardiol*, 2009; 53: 2070–7.
 25. Clerico A, Giannoni A, Vittorini S, Passino C. Thirty years of the heart as an endocrine organ: physiological role and clinical utility of cardiac natriuretic hormones. *Am J Physiol Heart Circ Physiol*, 2011; 301: H12–20.
 26. McCord J, Mundy BJ, Hudson MP, et al. Relationship between obesity and B-type natriuretic peptide levels. *Arch Intern Med.*, 2004; 164: 2247–52.
 27. Birkenfeld AL, Boschmann M, Moro C et al. Lipid mobilization with physiological atrial natriuretic peptide concentrations in humans. *J Clin Endocrinol Metab*, 2005; 90: 3622–3628.