10 best publications of Dr. Satinath Mukhopadhyay (with citations)

1. S Dasgupta, S Bhattacharya, A Biswas, S Majumdar, **S Mukhopadhyay**, S Bhattacharya. NF-kappaB mediates lipid-induced **fetuin-A** expression in hepatocytes that impairs adipocyte function effecting insulin resistance. **Biochem**. J 2011; 429, 451-462. Impact Factor: 4.654. **[Citations: 156]**

Summary: Fetuin-A, a hepatic secretory protein, has recently been implicated in insulin resistance and Type 2 diabetes. It is an endogenous inhibitor of insulin receptor tyrosine kinase. However, regulation of fetuin-A synthesis in relation to insulin resistance is unclear. In the present paper, we report that both non-esterified ('free') fatty acids and fetuin-A coexist at high levels in the serum of db/db mice, indicating an association between them. For an in-depth study, we incubated palmitate with HepG2 cells and rat primary hepatocytes, and found enhanced fetuin-A secretion to more than 4-fold over the control. Interestingly, cell lysates from these incubations showed overexpression and activity of NF-kappaB (nuclear factor kappaB). In NF-kappaB-knockout HepG2 cells, palmitate failed to increase fetuin-A secretion, whereas forced expression of NF-kappaB released fetuin-A massively in the absence of palmitate. Moreover, palmitate stimulated NF-kappaB binding to the fetuin-A promoter resulting in increased reporter activity. These results suggest NF-kappaB to be the mediator of the palmitate effect. Palmitate-induced robust expression of fetuin-A indicates the occurrence of additional targets, and we found that fetuin-A severely impaired adipocyte function leading to insulin resistance. Our results reveal a new dimension of lipid-induced insulin resistance and open another contemporary target for therapeutic intervention in Type 2 diabetes.

2. D Pal, S Dasgupta, R Kundu, S Maitra, G Das, **S Mukhopadhyay**, S Ray, S Bhattacharya. **Fetuin-A** acts as an endogenous ligand of TLR4 to promote lipid-induced insulin resistance. **Nature Medicine** 2012; 18 (8), 1279-1285. **Impact Factor: 22.864. [Citations: 744]**

Summary: Toll-like receptor 4 (TLR4) has a key role in innate immunity by activating an inflammatory signaling pathway. Free fatty acids (FFAs) stimulate adipose tissue inflammation through the TLR4 pathway, resulting in insulin resistance. However, current evidence suggests that FFAs do not directly bind to TLR4, but an endogenous ligand for TLR4 remains to be identified. Here we show that fetuin-A (FetA) could be this endogenous ligand and that it has a crucial role in regulating insulin sensitivity via Tlr4 signaling in mice. FetA (officially known as Ahsg) knockdown in mice with insulin resistance caused by a high-fat diet (HFD) resulted in downregulation of Tlr4-mediated inflammatory signaling in adipose tissue, whereas selective administration of FetA induced inflammatory signaling and insulin resistance. FFA-induced proinflammatory cytokine expression in adipocytes occurred only in the presence of both FetA and Tlr4; removing either of them prevented FFA-induced insulin resistance. We further found that FetA, through its terminal galactoside moiety, directly binds the residues of Leu100-Gly123 and Thr493-Thr516 in Tlr4. FFAs did not produce insulin resistance in adipocytes with mutated Tlr4 or galactoside-cleaved FetA. Taken together, our results suggest that FetA fulfills the requirement of an endogenous ligand for TLR4 through which lipids induce insulin resistance. This may

position FetA as a new therapeutic target for managing insulin resistance and type 2 diabetes.

3. P Chatterjee, S Seal, S Mukherjee, R Kundu, **S Mukhopadhyay**, S Ray, S Bhattacharya. Adipocyte **fetuin-a** contributes to macrophage migration into adipose tissue and polarization of macrophages. **Journal of Biological Chemistry** 2013; 288 (39), 28324- 28330.**Impact factor: 4.651 [Citations: 96]**

Summary: Macrophage infiltration into adipose tissue during obesity and their phenotypic conversion from anti-inflammatory M2 to proinflammatory M1 subtype significantly contributes to develop a link between inflammation and insulin resistance; signaling molecule(s) for these events, however, remains poorly understood. We demonstrate here that excess lipid in the adipose tissue environment may trigger one such signal. Adipose tissue from obese diabetic db/db mice, high fat diet-fed mice, and obese diabetic patients showed significantly elevated fetuin-A (FetA) levels in respect to their controls; partially hepatectomized high fat diet mice did not show noticeable alteration, indicating adipose tissue to be the source of this alteration. In adipocytes, fatty acid induces FetA gene and protein expressions, resulting in its copious release. We found that FetA could act as a chemoattractant for macrophages. To simulate lipid-induced inflammatory conditions when proinflammatory adipose tissue and macrophages create a niche of an altered microenvironment, we set up a transculture system of macrophages and adipocytes; the addition of fatty acid to adipocytes released FetA into the medium, which polarized M2 macrophages to M1. This was further confirmed by direct FetA addition to macrophages. Taken together, lipid-induced FetA from adipocytes is an efficient chemokine for macrophage migration and polarization. These findings open a new dimension for understanding obesity-induced inflammation.

4. **Mukhopadhyay S**, Mondal SA, Kumar M, Dutta D. Pro-inflammatory and anti-inflammatory attributes of **fetuin-A**: a novel hepatokine: modulating cardiovascular and glycemic outcomes in metabolic syndrome. Endocr Pract. 2014 Nov 4:1-18. **Impact factor 2.9. [Citations: 53]**

Summary: Fetuin-A is a novel hepatokine. The number of biologic roles attributed to fetuin-A has increased exponentially in the past decade. The objective of this review is to discuss the pathophysiology of fetuin-A action, its proinflammatory and antiinflammatory attributes in different biological systems throughout the body, and pharmacologic interventions that modulate fetuin-A levels.

PubMed, Medline, and Embase search for articles published to July 2014, using the terms "alpha-2-hs-glycoprotein" [MeSH Terms] OR "alpha-2-hs-glycoprotein" [All Fields] OR "fetuin a" [All Fields].

Fetuin-A is the endogenous ligand for Toll-like receptor-4 activation, for lipid-induced insulin resistance. Fetuin-A has inverse interaction with adiponectin. Increased fetuin-A is a risk factor for diabetes and fatty liver disease in normoglycemia and prediabetes. Fetuin-A is a negative acute-phase reactant in sepsis and endotoxemia, promotes wound

healing, and is neuroprotective in Alzheimer's disease. Decreased fetuin-A predicts increased disease activity in obstructive lung disease, Crohn's disease, and ulcerative colitis. Both elevated and reduced fetuin-A may be linked with increased cardiovascular events.

Fetuin-A is a pleotropic molecule with diverse (sometimes even contradictory) effects in different systems, brought about by interaction with a variety of receptors, including the insulin, transforming growth factor-β, and a plethora of Toll-like receptors. As a proinflammatory molecule, fetuin-A contributes to insulin resistance and is an important link between liver, adipose tissue, and muscles. Fetuin-A is neuroprotective and plays an important antiinflammatory role in sepsis and autoimmune disorders. Pharmacologic options are limited in modulating serum fetuin-A, but salsalates, curcumin, and vitamin D are promising agents of the future.

5. Agarwal S, Chattopadhyay M, Mukherjee S, Dasgupta S, **Mukhopadhyay S**, Bhattacharya S. **Fetuin-A** downregulates adiponectin through Wnt-PPARγ pathway in lipid induced inflamed adipocyte. **Biochim Biophys Acta**. 2016 Oct 6;1863(1):174-181. **[Citations: 21]**

Summary: Adiponectin secreted from adipocytes is an anti-diabetic and anti-atherogenic adipokine. Adiponectin level is known to fall significantly in obesity induced type 2 diabetes which worsen insulin sensitivity because of aberrant lipid management. However, underlying mechanism of adiponectin decrease in obese diabetic condition is yet unclear. We report here that lowering of plasma adiponectin coincided with the higher Fetuin A (FetA) level in high fat diet (HFD) induced obese diabetic mice. Knock down of FetA gene ($FetA^{KD}$) elevated adiponectin level markedly in HFD mice, while reinforcement of FetA into $FetA^{KD}$HFD mice reduced its level again. These results indicate FetA's involvement in the lowering of adiponectin level in obesity induced diabetic mice. Our findings to understand how FetA could affect adiponectin decrease demonstrated that FetA could enhance Wnt3a expression in the adipocyte of HFD mice. FetA addition to 3T3L1 adipocyte incubation elevated Wnt3a expression in a dose dependent manner. Overexpression of Wnt3a by FetA inhibited PPARγ and adiponectin. FetA failed to reduce PPARγ and adiponectin in Wnt3a gene knocked down 3T3L1` adipocytes. All these suggest that FetA mediate its inhibitory effect on adiponectin through Wnt3a-PPARγ pathway. Inhibition of adiponectin expression through FetA and Wnt3a significantly compromised with the activation of AMPK and its downstream signalling molecules which adversely affected lipid management causing loss of insulin sensitivity. Downregulation of adiponectin in inflamed adipocyte by FetA through the mediation of Wnt3a and PPARγ is a new report.

6. **Mukhopadhyay S**, Bhattacharya S. Plasma **fetuin-A** triggers inflammatory changes in macrophages and adipocytes by acting as an adaptor protein between NEFA and TLR- 4. **Diabetologia**. 2016 Apr;59(4):859-60. **[Citations: 15]**

Summary: We have shown that circulating NEFA stimulates the production of fetuin-A by the liver via NF-κB. Fetuin-A then forms a dimer with NEFA by acting as its

binding protein. The NEFA–fetuin-A dimer finally binds to TLR-4 present on the surface of adipocytes and macrophages, resulting in the formation of a ternary complex that triggers a local inflammatory response in the adipose tissue. Upon stimulation by circulating fetuin-A, white adipose tissue attracts circulating M2 macrophages that undergo proinflammatory polarisation to M1 under the influence of fetuin-A generated locally by the inflamed adipocytes.

7. Mukhuty A, Fouzder C, Mukherjee S, Malick C, **Mukhopadhyay S**, Bhattacharya S, Kundu R. Palmitate induced **Fetuin-A** secretion from pancreatic β-cells adversely affects its function and elicits inflammation. **Biochem Biophys Res Commun**. 2017 Sep 30; 491(4):1118-1124. **[Citations: 21]**

Summary: Islets of type 2 diabetes patients display inflammation, elevated levels of cytokines and macrophages. The master regulator of inflammation in the islets is free fatty acids (FFA). It has already been reported that FFA and TLR4 stimulation induces pro-inflammatory factors in the islets. In this report we demonstrate that excess lipid triggers Fetuin-A (FetA) secretion from the pancreatic β-cells. Palmitate treatment to MIN6 cells showed significantly elevated FetA levels in respect to their controls. Fatty acid induces the FetA gene and protein expression in the pancreatic β-cells via TLR4 and over-expression of NF-κB. In the NF-κB knocked down MIN6 cells palmitate could not trigger FetA release into the incubation medium. These results suggest that NF-κB mediates palmitate stimulated FetA secretion from the pancreatic β-cells. Blocking the activity of TLR4 by CLI-095 incubation or TLR4 siRNA restored insulin secretion which confirmed the role of TLR4 in FFA-FetA mediated pancreatic β-cell dysfunction. Palmitate mediated expression of NF-κB enahnced inflammatory response through expression of cytokines such as IL-1β and IL-6. These results suggest that FFA mediated FetA secretion from pancreatic β-cells lead to their dysfunction via FFA-TLR4 pathway. FetA thus creates an inflammatory environment in the pancreatic islets that can become a possible cause behind pancreatic β-cell dysfunction in chronic hyperlipidemic condition.

8. Mukherjee S, Chattopadhyay M, Bhattacharya S, Dasgupta S, Hussain S, Bharadwaj SK, Talukdar D, Usmani A, Pradhan BS, Majumdar SS, Chattopadhyay P, **Mukhopadhyay S**, Maity TK, Chaudhuri MK, Bhattacharya S. A Small Insulinomimetic Molecule Also Improves Insulin Sensitivity in Diabetic Mice. **PLoS One**. 2017 Jan 10;12(1):e0169809. **[Citations: 16]**

Summary: Dramatic increase of diabetes over the globe is in tandem with the increase in insulin requirement. This is because destruction and dysfunction of pancreatic β-cells are of common occurrence in both Type1 diabetes and Type2 diabetes, and insulin injection becomes a compulsion. Because of several problems associated with insulin injection, orally active insulin mimetic compounds would be ideal substitute. Here we report a small molecule, a peroxyvanadate compound i.e. DmpzH[$VO(O_2)_2$(dmpz)], henceforth referred as dmp, which specifically binds to insulin receptor with considerable affinity (KD-1.17μM) thus activating insulin receptor tyrosine kinase and its downstream signaling molecules resulting increased uptake of [^{14}C] 2 Deoxy-glucose. Oral administration of dmp to streptozotocin treated BALB/c mice lowers blood glucose level and markedly stimulates glucose and fatty acid uptake by skeletal muscle and adipose tissue respectively. In *db/db* mice, it greatly improves insulin

sensitivity through excess expression of PPARγ and its target genes i.e. adiponectin, CD36 and aP2. Study on the underlying mechanism demonstrated that excess expression of Wnt3a decreased PPARγ whereas dmp suppression of Wnt3a gene increased PPARγ expression which subsequently augmented adiponectin. Increased production of adiponectin in *db/db* mice due to dmp effected lowering of circulatory TG and FFA levels, activates AMPK in skeletal muscle and this stimulates mitochondrial biogenesis and bioenergetics. Decrease of lipid load along with increased mitochondrial activity greatly improves energy homeostasis which has been found to be correlated with the increased insulin sensitivity. The results obtained with dmp, therefore, strongly indicate that dmp could be a potential candidate for insulin replacement therapy.

9. D Meher, D Dutta, S Ghosh, P Mukhopadhyay, S Chowdhury, **S Mukhopadhyay**. Effect of a mixed meal on plasma lipids, insulin resistance and systemic inflammation in non- obese Indian adults with normal glucose tolerance and treatment naïve type-2 diabetes, **Diabetes research and clinical practice.** 2014; Apr;104(1):97-102. **Impact Factor: 2.741 [Citations: 13]**

Summary: Asian Indians are believed to have a lower capacity to clear a glucose load even during normoglycemia. High post meal glucose levels have been linked to postprandial dyslipidemia and generation of proinflammatory cytokines. Since humans spend most of their time in the postprandial state, the present study aims to evaluate the relationship of insulin resistance (IR) in the basal state with dyslipidemia and systemic inflammation (hs-CRP, IL-6 and TNF-a), in the fasting state, 2h and 4h after a mixed meal, in Indian adults with normal glucose tolerance, and new onset type-2 diabetes.

Forty-eight people with type 2 diabetes and 32 individuals with normoglycemia, 30-70 years age, not on medications, underwent blood sampling after overnight (12h) fast and 2 and 4h after a mixed meal (carbohydrates, proteins and fat content 79.1%, 7.7% and 13.2%, respectively).

Triglyceride (TG), TG/HDL-C (high density lipoprotein), HDL-C/LDL-C (low density lipoprotein) ratios, IR parameters, and inflammatory markers were significantly higher among patients with diabetes. There was a fall in total cholesterol (TC), HDL-C and LDL-C at 2 and 4h after the meal in both groups. Compared with fasting, 4-h postprandial TC, TG and HDL-C were significantly better positively correlated with IR in normal individuals. Postprandial hs-CRP was not significantly different to fasting in both groups. Postprandial IL-6 and TNF-α were significantly lower in both groups. Consumption of a carbohydrate rich meal is associated with a rise in TG and fall in TC, HDL-C, LDL-C, IL-6 and TNF-α among normal individuals and people with type 2 diabetes.

10. D Dutta, S Choudhuri, SA Mondal, I Maisnam, AHH Reza, S Ghosh, **S Mukhopadhyay**. Tumor necrosis factor alpha− 238G/A (rs 361525) gene polymorphism predicts progression to type-2 diabetes in an Eastern Indian population with prediabetes. **Diabetes research and clinical practice.** 2013; Mar;99(3):e37-41. **Impact factor: 2.741 [Citations: 25]**

Summary: Prediabetes (IPD; n=122) and normoglycemic individuals (n=100) underwent assessment of polymorphisms of TNFα (-238, -308) and IL6 (-174). After 27.25±5.64 months, 16 IPD had reverted to normoglycemia and 18 progressed to diabetes. TNFα -238AA/GA genotypes were significantly more common in IPD, had higher TNFα, higher progression to diabetes and lower reversal.