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Toll-Like Receptor Activation and B Cell Maturation Via MyD88-Dependent Pathway Under Hyperglycemia Condition

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Abstract *–Hyperglycemia causes degenerative syndrome that involves an inflammatory process with an increase in certain proinflammatory cytokines and chemokines which in the process will activate B cells to produce immunoglobulins through several mechanisms. One of the interesting mechanisms is mechanism via MyD88 pathway. This review aimed to explore the role of MyD88 adapter protein in Toll like receptor activation and B cell maturation under hyperglycemia condition. A literature review was done to answer the study objectives by selecting related articles published in Google Scholar and Pubmed from 2013-2023. Out of 369 articles screened for eligibility and 31 studies included in the analysis. Class switching process under hyperglycemia condition involves activation of NFkB through the inflammatory MyD88-dependent pathway to trigger the expression of TLR and B cell maturation and proliferation as well as antibody production. The MyD88 adapter protein is a protein formed by stimulation of pro-inflammatory cytokines IL-6 and plays a role in the continuation of signals from the TLR and IL-1 pathways. Mature B cell stimulation induces 2 genetic changes in the Ig gene locus, called somatic hypermutation (SHM) and class switch recombination (CSR) to produce antibodies. Changes in immunoglobulin genes occur related to changes in certain DNA segments at the locus of genes where CSR occurs, this gene change requires the role of AID (activated-induced cytidine deaminase) in DNA cleavage. AID in mature B cells is activated by proinflammatory cytokines via induction of NfKB activation via the inflammatory MyD88-dependent pathway. Toll-like receptor activation plays a crucial role in B cell maturation activated by pro inflammatory cytokine via MyD88 dependent-NFkB activation.*

Keywords: *proinflammatory cytokines, TLR, B cell maturation, MyD88, hyperglicemia*

INTRODUCTION

Hyperglycemia originates from the Greek words hyper (meaning high), glykys (meaning sugar), and haima (meaning blood), and it refers to elevated levels of sugar in the blood (Mouri & Badireddy, 2023). Hyperglycemia refers to an elevation in blood glucose levels over 140 mg/dl (Korytkowski et al., 2012; Pasquel et al., 2021).

The prevalence of hyperglycemia ranges from 3.2% to 20% (Barcelo et al., 2012; Gebreyes et al., 2018). The fluctuating incidence is determined by numerous factors, including age and place of residency (Lai et al., 2000; Quang Binh et al., 2012). Getting older promotes hyperglycemia in both animal studies and humans (Chia et al., 2018; Ham et al., 2019). This phenomenon arises from a reduction in the capacity of pancreatic β-cells to proliferate, a decline in insulin release, and an elevation in insulin resistance (Ham et al., 2019; Lee & Halter, 2017) This condition is worsened by the presence of comorbid factors such as hypertension, inflammation, use of drugs that increase blood sugar levels (Lee & Halter, 2017). Populations residing in green spaces generally have a reduced likelihood of experiencing hyperglycemia, whereas communities residing in areas characterized by crime, air pollution, and noise tend to have an increased risk of hyperglycemia (Dendup et al., 2018; Kolb & Martin, 2017).

Hyperglycemia can manifest either suddenly or persistently. Acute hyperglycemia applies to a transient elevation in blood glucose levels over 140 mg/dl (Argyropoulos et al., 2021; Pasquel et al., 2021). Acute hyperglycemia commonly arises

in settings of heightened glucose levels, such as preoperative states, severe infections like COVID-19, febrile infections and convulsions, severe acute illnesses, trauma, and burns (Argyropoulos et al., 2021; Gojda et al., 2023). Chronic hyperglycemia commonly occurs in individuals experiencing chronic stress, obesity, pancreatitis, and diabetes mellitus. Diabetes mellitus (DM) is the primary cause of chronic hyperglycemia (Angeli et al., 2015; Duan et al., 2014; Nouhjah et al., 2017).

Hyperglycemia induces microvascular anomalies that can cause degenerative syndromes, which in turn contribute to the development and advancement of diseases characterized by inflammatory processes. This process is marked by an augmented production of proinflammatory cytokines and chemokines (Li et al., 2023; Nedosugova et al., 2022). The proinflammatory molecules will stimulate the proliferation and differentiation of B cells, leading to the generation of immunoglobulins through multiple pathways, including the MyD88-dependent pathway (Uchiyama et al., 2015; Vazquez et al., 2015).

MyD88 was originally identified as a protein that is produced in response to IL-6 stimulation during the last stage of maturation of M1 myeloleukemia cells (Chen et al., 2020). It is now understood that MyD88 also functions in the transmission of signals from the TLR and IL-1 pathways (Warner & Núñez, 2013). The reason for this is the fact that MyD88 possesses a distinctive modular arrangement, comprising an inactive domain at the N terminus that bears resemblance to the intracellular segment of TNF 1 and Fas receptors, and a C terminal portion that is similar to the cytoplasmic domain of Toll in *Drosophila* and the interleukin-1 receptor in vertebrates (Chen et al., 2020). Consequently, it can be inferred that MyD88 plays a pivotal function in the innate immune system (Friedrich et al., 2017). The aim of this study was to ascertain the function of the MyD88 adapter protein in the pathways of TLR activation and B cell maturation under hyperglycemic conditions.

MATERIALS AND METHODS

This work is a literature review that utilized a literature search methodology and underwent reviewed by two experts. This study utilized literature search using the terms toll-like receptors, B cell maturation, MyD88, and hyperglycemia. Articles with duplicate titles, discrepancies in concepts, background, and scope of discussion, as well as those which failed to provide a fulltext manuscript available, were excluded. A total of 369 were identified based on the articles search result and final evaluation was determined that 31 studies were included for analysis. Illustration of articles screening is shown in Figure 1.

RESULT AND DISCUSSION

1. Activation of Toll-Like Receptors (TLRs)

TLR activation originates as a result of its interaction with pathogen associated molecular patterns (PAMP). This process entails the binding of diverse adaptor proteins, thereby leading to the activation of multiple protein kinases. An active protein kinase initiates the activation of nuclear factor κappa B (NFκB) and interferon related factor 3 (IRF-3), leading to the activation of multiple genes, including those that encode inflammatory cytokines (tumor necrozing factor α / TNF α, interleukin 1 / IL-1, IL-2), chemokines (IL-8, MCP-1, RANTES), and antiviral cytokines (IFN-α/β). TLR signals comprise both MyD88-dependent and MyD88-independent pathways (De Oliveira et al., 2019; Federico et al., 2020; Jatmiko & Aisyah, 2015).

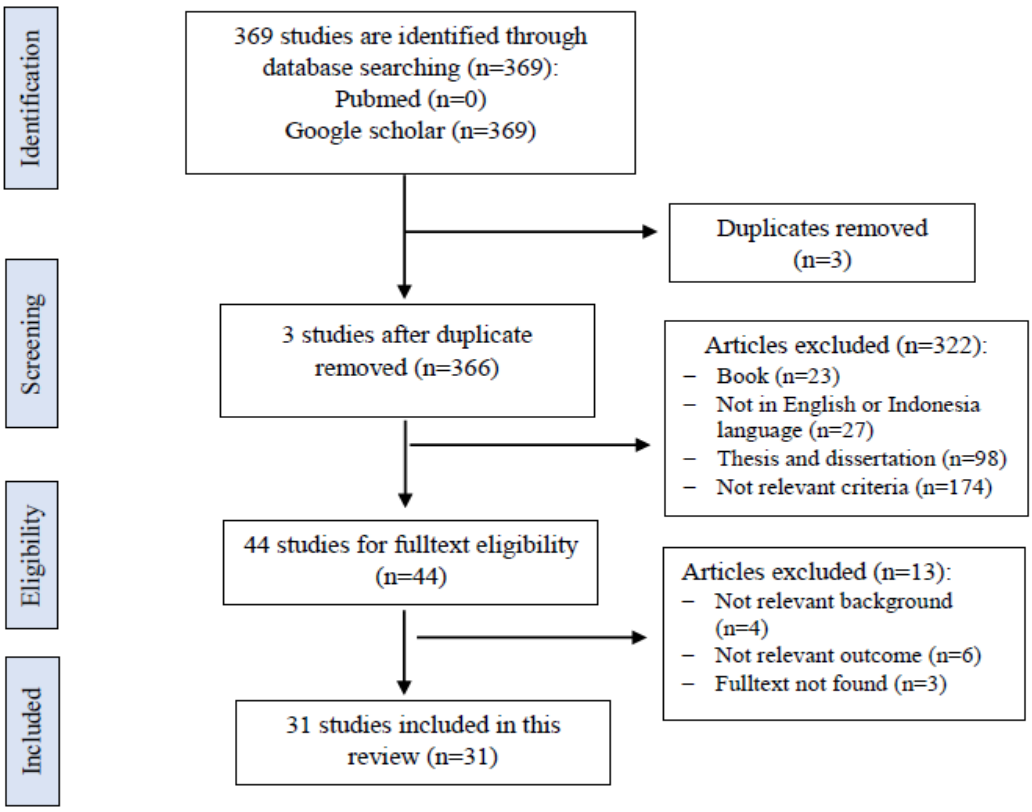

Figure 1. PRISMA diagram for included studies

The MyD88-dependent pathway is initiated by the binding of MyD88 to the TIR domain of the TLR or the association of MyD88 with MAL, which is already bound to the TLR. Subsequently, MyD 88 will enlist IRAK 4 (IL-1 receptor-associated kinase 4) and IRAK 1. Recruitment leads to the activation of IRAK 4. IRAK 4 induces excessive phosphorylation of IRAK 1. The outcome of hyperphosphorylation of IRAK 1 is the formation of a complex involving TRAF 6 (TNF receptor-associated factors 6), resulting in the IRAK4-IRAK1-TRAF6 complex. The incorporation of TRAF6 into the complex induces structural alterations within the complex, leading to the liberation of IRAK4-IRAK1-TRAF6. At that point, the IRAK4-IRAK1-TRAF6 complex forms a binding interaction with the pre-existing TAK1, TAB1, and TAB2 complexes located in the cell membrane. As a consequence of this interaction, TAB2 and TAK1 undergo phosphorylation. Furthermore, TAB2, TAK1, TAB1, and TRAF6 are liberated into the cytosol. TAK1 is activated within the cytosol, enabling it to activate IKK, which is an inhibitor of NF-κB kinase. IKK is activated and causes the degradation of IκB, which is an inhibitory protein. This degradation leads to the release of NFκB. In addition, TAK1 also triggers the activation of MAPK (mitogen-activated protein kinases) and JNK (JUN N terminal kinase) (Faghfouri et al., 2020; Jatmiko, 2018; Jin et al., 2013; Jin & Flavell, 2013; Krishnan-Sivadoss et al., 2021; Needell & Zipris, 2017; Sepehri et al., 2016; Velloso et al., 2015; Z. Wang et al., 2020; Xu et al., 2019; Yin et al., 2013).

2. Hyperglycemia elevates the expression of TLR

Hyperglycemia upregulates the expression of Toll-like receptor 2 (TLR-2)

and Toll-like receptor 4 (TLR-4). The upregulation of TLR-2 and TLR-4 expression is concomitant with an elevation in ROS, Nox, and PKC levels. Hyperglycemia triggers the activation of PKC-α and PKC-δ, leading to the activation of NADPH oxidase (Nox). Nox activation will result in the generation of superoxide. In addition, superoxide enhances the expression of TLR-2 and TLR-4. Hyperglycemia enhances the expression of TLR-2 and TLR-4, as well as other TLRs. The impact of hyperglycemia on the increase of TLR expression is transient. The duration of this impact will cease during a span of 48 hours, when blood glucose levels revert back to their usual state (Aghamiri et al., 2022; Dasu & Martin, 2014; Liu et al., 2014; Lucas & Maes, 2013; Peng et al., 2017; Rastogi et al., 2017; Santoni et al., 2015; Wang, et al., 2020).

3. Hyperglycemia promotes TLR dimerization

Hyperglycemia circumstances not only enhance the expression of TLR, but also initiate the formation of TLR dimers. The process of TLR dimerization involves the formation of both homodimers and heterodimers. The process of homodimer formation is characterized by the homodimerization of TLR-4, whereas heterodimerization is achieved through the dimerization of TLR-2 with TLR-6. TLR dimerization is not limited to TLR-2, TLR-4, and TLR-6, but is also possible in other TLRs (Dasu & Martin, 2014; Kochumon et al., 2022; Sepehri et al., 2016).

4. Hyperglycemia enhances Myd88 expression

Hyperglycemia enhances the expression of adaptor proteins necessary for transmitting signals within cells. Two adaptor proteins, MyD88 and TRIF, exhibit increased expression in the presence of high blood sugar levels (hyperglycemia). These two adapter proteins are essential for transmitting signals into cells upon activation of TLR. This enhances and augments cell activation via TLR by utilizing both the MyD88-dependent and MyD88-independent pathways (Liu et al., 2014; Luo et al., 2021; Santoni et al., 2015; L. Wang et al., 2015b; Ye & Steinle, 2016).

5. Hyperglycemia activates TLRs and Increases Proinflammatory cytokine Secretion via the Myd88 Pathway

There are plenty of ligands that interact with Toll-like receptors (TLRs). These include high mobility group Box 1 (HMGB1), heat shock proteins (HSP), lipopolysaccharide (LPS), and reactive oxygen species (ROS). Hyperglycemia conditions lead to a rise in HMGB-1 and ROS (Mudaliar et al., 2014; Ning et al., 2022; Wu et al., 2016).

ROS can alter the components of the cell membrane and release substances that can interact with and activate TLRs. This is reinforced by a rise in NF-κB and AP-1 following the activation of TLR-2 by ROS (Fuentes-Antrás et al., 2014). HMGB-1 is released from cells by two distinct mechanisms: actively, in response to oxidative stress, and passively, when cells undergo necrosis (Wu et al., 2016). HMGB-1 activates TLRs (Berger et al., 2016; Nogueira-Machado, 2011). The production of pro-inflammatory cytokines is triggered by the role of NF-κB on the activation of pro-inflammatory cytokine genes (Westwell-Roper et al., 2014; Rogero & Calder, 2018; X. Wang et al., 2020; Ye & Steinle, 2016).

6. Proinflammatory Molecular Pathway by Myd88 Triggers Immunoglobulin Class Switching

Previous studies has demonstrated that elevated glucose levels lead to an upregulation of TLR-4, MyD88, and IL-1β expression.

This suggests that hyperglycemia triggers the production and activation of TLR4 in endothelial cells (L. Wang et al., 2015a). All Toll-like receptors (TLRs) except TLR3 utilize the MyD88 adapter protein signaling pathway (Lannoy, Côté-biron, et al., 2023). Additionally, TLRs and MyD88 together control the expression of proinflammatory cytokines (Kiripolsky et al., 2020). A further study demonstrated that the inflammatory capacity of the Vi vaccine relies on TLR-MyD88, a crucial factor in the generation of class-switched IgG antibodies (Garg et al., 2015). The findings indicate that proinflammatory molecular pathways enhance the occurrence of class switch recombination (CSR) of immunoglobulin genes in mature B cells (Corsiero et al., 2016).

Recent studies indicate that the presence of high sugar levels leads to an elevation in the expression of TLR-4, MyD88, and IL-1β (Lannoy et al., 2023; Wada & Makino, 2016). This suggests that hyperglycemia stimulates the production of TLR4 in endothelial cells. Hyperglycemia is a prevalent degenerative condition with multiple contributing factors (Mouri & Badireddy, 2023). Elevated blood sugar levels can initiate an inflammatory response and lead to vascular problems associated with inflammation (Li et al., 2023; Morris, 2015; Nedosugova et al., 2022). Endothelial cells exhibit various inflammatory reactions when exposed to high glucose, including activation of NFkB (Suryavanshi & Kulkarni, 2017), reduced production of nitric oxide (Meza et al., 2019), increased expression of inflammatory genes (Wang et al., 2015), recruitment of leukocytes and activation of T cells due to enhanced production of chemoattractants (Suryavanshi & Kulkarni, 2017), and promotion of B cell proliferation, differentiation, and antibody production (Szukiewicz, 2023; Zhai et al., 2016). The immune response is initiated by activating NFκB through the MyD88-dependent inflammatory pathway, which leads to the expression of TLR (Dasu & Martin, 2014) and the maturation, proliferation, and generation of antibodies in B cells (Yehualashet, 2020; Zhai et al., 2016).

Activation of these fully developed B cells triggers two genetic modifications in the Ig gene locus, identified as somatic hypermutation (SHM) and class switch recombination (CSR), which generate antibodies with higher affinity and diverse effector capabilities. CSR in the DNA segment induces a modification in the CH gene for IgM, leading to the substitution with one of the CH genes for IgG, IgE, or IgA. This alteration aids in the elimination of antigens. Mature B cells possess a fully functional variable recombination, diversity, joining (VDJ) function in both heavy chain (H chain) and light chain (L chain) genes (Chi et al., 2020). They display IgM on their surface and move to secondary lymphoid organs. In these organs, the IgM interacts with antigens, leading to the activation of B cells (Althwaiqeb & Bordoni, 2023).

B cells that are currently activated will initiate class switch recombination (CSR) and somatic hypermutation (SHM) processes at the immunoglobulin (Ig) gene locus, while maintaining their antigen specificity. Activation-induced cytidine deaminase (AID) is a crucial enzyme for DNA cleavage in class switch recombination (CSR) of the immunoglobulin (Ig) gene, playing a vital role in the CSR mechanism. AID is especially present on activated B cells, which play a crucial role in initiating the DNA cleavage phase in both class switch recombination (CSR) and somatic hypermutation (SHM). Furthermore, the proper functioning of CSR in Ig genes necessitates not only the crucial involvement of AID, but also the induction of protein production by RNA editing and

the formation of AID-RNA complexes (Chi et al., 2020).

CONCLUSION

Hyperglycemia conditions lead to a rise in ROS which can alter the omponents of the cell membrane and release substances that can interact with and activate TLRs. All Toll-like receptors (TLRs) except TLR3 utilize the MyD88 adapter protein signaling pathway. Additionally, TLRs and MyD88 together control the expression of proinflammatory cytokines. TLR-MyD88 is a crucial factor in the generation of class-switched IgG antibodies. Poinflammatory molecular pathways enhance the occurrence of CSR of immunoglobulin genes in mature B cells.

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