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Voltage dependent anion channel-mediated apoptosis: Its role in the pathogenesis of diabetic complications

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Abstract

Apoptosis or programme cell death is a normal phenomenon required for maintaining cell homeostasis. Several studies indicate that excessive or insufficient apoptosis can lead to various diseases. In diabetes enhanced cellular apoptosis has been associated with micro and macro-vascular complications. Accumulating evidences suggest the involvement of voltage-dependent anion channel (VDAC) towards increased apoptosis observed in diabetes. VDAC, a multifunctional mitochondrial porin is a protein located on the outer mitochondrial membrane which contributes to apoptosis either through Ca^{2+} overload into mitochondria or by release of apoptotic protein from the mitochondria by opening the mitochondrial permeability transition pore (mPTP) or rupture of the mitochondrial outer membrane. Here, we provide a short review on VDAC-mediated apoptosis and its pathogenic role in the progression of diabetic complications.

Keywords: Apoptosis, diabetes, voltage-dependent anion channel, diabetic complications.

Introduction

Diabetes is a metabolic disorder characterized by hyperglycemic condition caused by deficiency in the secretion of insulin, ineffective insulin action or both (American Diabetes Association, 2009). There are two major forms of diabetes: type 1 (T1D) and type 2 (T2D). T1D results from insufficient insulin production due to destruction of β cells of the pancreas (American Diabetes Association, 2009). T2D results from impaired insulin secretion and insulin resistance and is associated with obesity, lack of exercise, stress, as well as aging. In both types of diabetes, the late diabetic complications in different tissues arise from chronic or intermittent hyperglycemia (Forbes and Cooper, 2013). Hyperglycemia-induced cell death due to apoptosis appears to play a pivotal role in micro-and macro-vascular complications such as angiopathy, retinopathy, neuropathy, atherosclerosis, impaired wound healing and periodontitis (Allen *et al.,* 2005). The present review will discuss the role of cellular death in diabetes with particular emphasis on apoptosis-mediated *via* voltage-dependent anion channel (VDAC) in the pathogenesis of diabetes as a disease.

Apoptosis in diabetes

Apoptosis or programme cell death is a coordinated series of events which is tightly regulated and occurs as a homeostatic mechanism to maintain cell population in tissues (Lee and Pervaiz, 2007). Apoptosis also occurs as a defence mechanism such as in immune reactions or when cells are damaged by disease or noxious agents. However, when apoptosis malfunctions, it results into a variety of pathological states where failure in apoptosis could lead to cancer, autoimmune diseases while excessive apoptosis could lead to cell loss such as in HIV/AIDS, neurodegenerative diseases and diabetes mellitus (Lee and Pervaiz, 2007). There are many mechanisms and pathways that can initiate apoptosis but the two distinct pathways are (i) extrinsic or death receptor pathway and (ii) intrinsic or mitochondria-mediated pathway as shown in Figure 1.

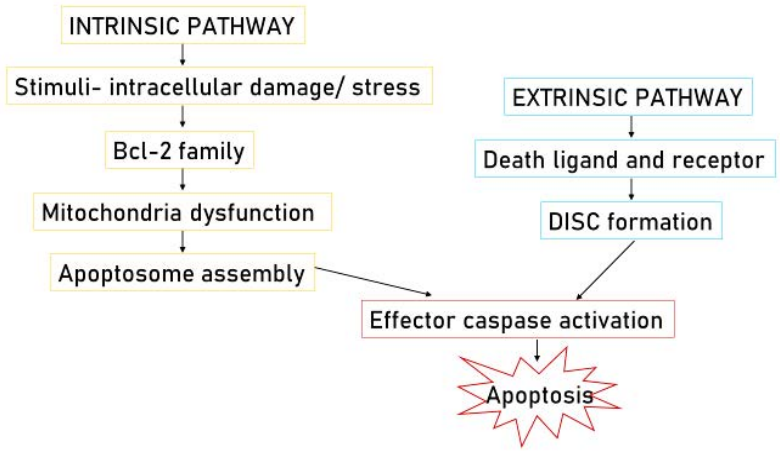

Figure 1. The intrinsic and extrinsic pathway of apoptosis. Apoptosis can occur through either one of the two pathways: extrinsic (death ligand and receptor mediated which involve TNF, Fas as some of the ligands with formation of death-inducing signalling complex, DISC being a critical step in Fas-mediated apoptosis). The latter pathway results from mitochondrial dysfunction which releases cytochrome *c* and subsequent activation of the caspases.

Extrinsic pathway is triggered following signals through death receptors such as tumour necrosis factor (TNF) family death receptors; the intrinsic pathway is activated by cellular stress such as oxidative stress (Zhaoyu and El-Deiry, 2005), Ca^{2+} overload and is regulated primarily by the pro-apoptotic and the anti-apoptotic members of the Bcl-2 family. Both pathways activate a cascade of proteolytic enzymes called caspases that mediate the rapid dismantling of cellular organelles and architecture although caspase-independent pathways have also been reported (Leist and Jaattela, 2001).

Several reports suggest the occurrence of enhanced apoptosis during diabetes i.e., in both T1D and T2D. Among which, apoptosis of β cells has been discussed most widely under

diabetes (Wali *et al.*, 2013). In T1D, β-cells are destroyed by immunological mechanisms, whereas in T2D metabolic abnormalities contribute to β-cell failure and subsequent apoptosis. Therefore, regardless of the diabetes type, highly conserved intracellular pathways of apoptosis are triggered. In T1D and T2D, β -cell death occurs by both the extrinsic and intrinsic pathway (Cnop *et al.,* 2005). Similarly, in other cells/tissues, both the pathways have been found to play a role under diabetes. Reports of apoptosis in other cells/tissues such as renal (Habib, 2013), hepatocytes (Schattenberg and Schuchmann, 2009), cardiomyocyte (Ouyang *et al.*, 2014), retina cells (Barber *et al.*, 2011) have been shown to cause diabetic complications such as diabetic nephropathy, diabetic neuropathy, liver injury, diabetic cardiomyopathy, diabetic retinopathy respectively (Feenstra *et al.*, 2013). The mechanisms that lead to diabetes-induced cell death are complex and are not fully understood but several lines of evidences have implicated that high glucose promotes the upregulation or activation of several molecules involved in different pro-apoptotic pathways and there are no specific pathways for high glucose-induced cell death. Many studies have demonstrated hyperglycemia-induced or glucose-induced intrinsic apoptosis in beta-cells (McKenzie *et al.,* 2010; Wali *et al.*, 2013) endothelial cells (Peng *et al.,* 2013), mesangial cells (Mishra *et al.,* 2005) etc. In intrinsic mitochondria apoptosis pathway cytochrome *c* is released from the intermembrane space of mitochondria through a process called mitochondrial outer membrane permeabilization (MOMP). MOMP is regulated by Bcl-2 protein family. The balance of pro- and anti-apoptotic Bcl-2 proteins in mitochondriał and endoplasmic reticulum membranes regulates MOMP in part by regulating calcium compartmentalization. Bcl-2, Bcl-XL and Mcl-1 prevent MOMP whereas pro-apoptotic members, Bax and Bak activate MOMP (Kroemer *et al.*, 2007). Once released, cytochrome *c* binds Apaf-1 (Apoptotic proteases activating factor), dATP and procaspase-9 creating a complex called apoptosome which activates procaspase-9 to cleave into caspase-9 generating caspase-3 eventually causing apoptosis. MOMP can also be initiated by an abrupt increase in the permeability to ions and small solutes of the inner mitochondrial membrane (IMM) by a process known as mitochondrial permeability transition (MPT). MPT appears to be mediated by a multiprotein complex that is assembled at the juxtaposition sites between the outer mitochondria membrane (OMM) and inner membrane (IMM), the so-called permeability transition pore complex (PTPC) (Kroemer *et al.*, 2007). One of the main components of the PTPC, is the mitochondrial voltage-dependent anion channel (VDAC) (Kroemer *et al.*, 2007) also known as mitochondrial porin which is the most abundant protein in OMM. Several studies have reported that VDAC is associated with T2D (Turko and Murad, 2003; Mostyn *et al.,* 2004) which is an important link between diabetes and mitochondrial function. VDAC has also been found to play a role in coronary endothelial cell dysfunction in T1D mice (Sepassi *et al.,* 2013). Thus, VDAC might contribute to apoptosis related to diabetes and is consequently the main focus of this review.

VDAC and its isoforms

VDAC acts as a channel for the transfer of ions and other small metabolites from outside into the mitochondria and as a selective channel for cations and uncharged molecules. In its open state, it allows the passage of hydrophilic molecules of oxidative phosphorylation substrates (pyruvate, oxaloacetate, malate, succinate, ATP, ADP, inorganic phosphate), urea cycle substrates and exchange of methyl groups (Colombini, 2004; Vander Heiden *et al.,* 2000). However, under special conditions these channels can be closed which reduces the permeability of OMM thereby, blocking metabolite exchange (Colombini, 2004; Holmuhamedov and Lemasters, 2009). VDAC exist as three isoforms VDAC1, 2 and 3 (Caterino *et al.,* 2017) and their amino acid sequences has already been determined. The three proteins have similar structure, conductance (Shoshan-Barmatz *et al.*, 2017), voltage-gating properties with molecular weights of 30-35 kDa and shares approximately 70% identity. All three can be found in most tissues with VDAC1 being the most abundant and VDAC3 being the least common form. All the three isoforms are encoded by distinct genes located on different chromosomes and they share the same exon-intron organization (De Pinto *et al.,* 2010). These VDACs complement each other and have some functional redundancy (Teplova *et al.*, 2011) although, they are significantly different with relation to functionality (Shoshan-Barmatz *et al.,* 2010; De Pinto *et al.,* 2010; De Pinto *et al.,* 2016). Table 1 shows the function of different VDAC isoforms.

Table 1. VDAC isoforms and functions

VDAC isoforms	VDAC function	References
VDAC1	Form oligomeric pores associated with stress and mitochondria DNA (mtDNA) release in diseases	Kim *et al.,* 2019
	Involved in transmission of Ca^{2+} signals to mitochondria during apoptosis	De Stefani *et al.,* 2011; Shoshan-Barmatz *et al.,* 2018
	Important for ROS control	Reina *et al.,* 2010
	Regulator of mitochondrial membrane permeabilization and apoptosis	Tajeddine *et al.,* 2008; Yuan *et al.,* 2008
	Interacts with hexokinase and Bcl-2 to mediate anti-apoptotic activity	Abu-Hamad *et al.,* 2009, Zhang *et al.,* 2019b

VDAC2	Important for ROS control	Reina *et al.,* 2010
	Abundant in the outer dense fiber and might be involved in the regulation of sperm motility	Hinsch *et al.,* 2004
	Porin2 isoform in *Drosophila melanogaster* suggested to be involved in sperm maturation	Guarino *et al.,* 2006
VDAC3	Possible involvement in mtDNA through formation of oligomeric pores	Kim *et al.,* 2019
	Abundant at the outer dense fiber of sperm flagellum and might be involved in the regulation of sperm motility	Hinsch *et al.,* 2004
	Deficient VDAC3 produces healthy infertile mice maybe due to immotile sperm	Sampson *et al.,* 2001
	Functional alteration of complex IV of the heart	Anflous-Pharayra *et al.,* 2011
	Moderates centriole assembly and recruits Msp1 protein to centrosomes	Majumder *et al.,* 2012

Structurally VDACs (primarily VDAC1) are β-barrel-forming transmembrane channels and through biochemical studies, it was shown to composed of one helix and 13 β-sheets with some relatively long loops between β-sheets which was proposed to play a role in protein-protein interaction (Colombini, 2004). However, crystal structure showed a β-barrel is formed by 19 β-sheets with the N-terminal α-helix lying within the pore (Ujwal *et al.,* 2008). Several studies suggest the N-terminal region mobility is involved in channel gating and serve as the interaction site of apoptosis-regulating proteins of the Bcl-2 family (Abu-Hamad *et al.,* 2009; Arbel *et al.*, 2012) and hexokinase (Arzoine, 2008). The N-terminal segment was also proposed to regulate cytochrome *c* release and subsequent apoptosis (Abu-Hamad *et al.,* 2009).

Role of VDAC-mediated apoptosis in diabetic complications

Chronic hyperglycemia and associated risk factors lead to irreversible diabetic complications affecting kidney, retina, peripheral nerves, brain and cardiovascular system. Diabetes mellitus both type 1 and type 2 are associated with an enhanced apoptosis of different cells and tissues which accelerates the occurrence of diabetic complications. Studies have shown that high glucose-mediated apoptotic cell death is relevant to diabetic complications as high glucose causes activation of several proteins involved in the apoptotic cell death (Allen *et al.*, 2005). There are several reports which have explained the possible mechanism by which high glucose increases the rate of apoptosis such as high glucose induced oxidative and nitrosative stress, *via* MAPK, p53 including mitochondria-dependent and-independent mechanisms particularly involving Bax or Bad. In previous

reports, high glucose in mesangial cells was shown to initiate oxidative-stress-induced apoptosis *via* Bax-mediated mitochondrial permeability and cytochrome *c* release (Allen *et al.,* 2005). In a process carried out by insulin-like growth factor-I (IGF-I), high glucose-induced apoptosis was prevented by IGF-I causing phosphorylation of Bad at Ser^{112}(Kang *et al.,* 2003).

Previous studies have demonstrated that high glucose exposure of pancreatic β-cells results into cell dysfunction and cell death. One of the possible factors as described by the study performed by Kim *et al*.(2005), is through decreasing glucokinase (GCK) or hexokinase IV protein expression and its interactions with VDAC which correlates with decrease in Bad phosphorylation. GCK belongs to the hexokinase family that converts glucose to glucose-6-phosphate by transferring a phosphate group from ATP to glucose, the initial step of glucose metabolism (Nordlie *et al.*, 1999). Glucose regulates Bad phosphorylation (Danial *et al.,* 2003) and phosphorylated Bad promotes interactions between hexokinase and VDAC necessary for pumping of ATP from mitochondria (Majewski *et al.,* 2004). Decreased Bad phosphorylation increases the chances of apoptosis. Bad is a member of the Bcl-2 family which induces apoptosis by inhibiting antiapoptotic Bcl-2 family members Bcl-xL, Bcl-2, allowing, Bak and Bax, pro-apoptotic proteins to aggregate and induce release of cytochrome *c* and eventually to apoptosis (Bergmann, 2002). High glucose exposure decreases the association of GCK to mitochondria, thereby increasing the interaction between Bax and mitochondria eventually leading to Bax oligomerisation, cytochrome *c* release followed by β-cell apoptosis (Kim *et al.,* 2005). Zhang *et al.* (2019a) reported that VDAC1 overexpression in T2D causes ATP loss in β- cell which on direct inhibition of VDAC1 restored glucose-stimulated insulin secretion (GSIS) and prevented development of diabetes in db/db mice. Study performed by Ahmed *et al.* (2010), on prolonged exposure of INS1E cell lines to high glucose was accompanied with marked expression of the VDAC1 and a reduction of VDAC2 suggesting that VDAC1 and VDAC2 expression could represent a consequence of early step in the β-cell dysfunction that could be an important target process to prevent altered insulin secretion and β-cell apoptosis. In other studies, hyperglycemia have shown to increase VDAC1 expression in β-cells (Salehi, 2010), in kidney (Gong *et al.,* 2009) including mouse coronary endothelial cells (MCECs) isolated from diabetic mice showed increased VDAC expression. VDAC inhibition restored the increased Ca^{2+}, O^{2-} and mPTP opening activity in diabetic MCECs suggesting normalising VDAC protein may decrease the incidence of cardiac ischemia in diabetes by decreasing endothelial cell apoptosis and increase in capillary density in the hearts (Sasaki *et al.,* 2012). Li *et al.* (2016) reported that IncRNA H19/miR-675 axis regulates cardiomyocyte apoptosis by targeting VDAC1 in diabetic cardiomyopathy. In retina, VDAC is localised to mitochondria predominantly at photoreceptors where results suggest that VDAC is involved in the regulation of cytosolic and mitochondrial Ca^{2+} concentration and mPTP assembly and/or activation. In retinal neurons especially ganglion cells, ischemia, diabetes apoptotic events appear to be promoted by an abnormal concentration of intracellular Ca^{2+}. VDAC being permeable to Ca^{2+} and as a component of mPTP may be involved in inducing mitochondrial swelling.

Therefore, VDAC may play a key role in processes of retinal degeneration that result from ATP depletion and Ca^{2+} overload. In one of the previous studies performed by Jones *et al.,* 2008 showed VDAC as a potential target of O-linked beta-N-acetylglucosamine (O-GlcNAc) modification. O-GlcNAc modification is a type of post-translation modification that modulates cellular function. In this study, they found that the adult mouse heart with elevated O-GlcNAc levels had more O-GlcNAc-modified VDAC and were found to be more resistant to calcium-induced swelling which was critical for survival providing a mechanism of cardioprotection. Increased apoptosis occurred in the tubular cells of db/db mice which on Mito Q administration were inhibited. Mito Q is a mitochondria-targeted antioxidant comprising of coenzyme Q10 and TPP cations. VDAC expression was found to increase in tubular of db/db mice.

However, there are contradictory results showing decreased VDAC on the mitochondrial kidneys of diabetic rats where the up-regulation of VDAC on α-Lipoic acid treatment exerted a protective role against mitochondrial injury (Wang *et al.*, 2013). In another study involving Zucker diabetic fatty obese (ZDFO) rats with early T2D, VDAC protein expression were significantly lower in the cerebral microvessels indicating deterioration of mitochondrial function in the cerebral vasculature during at an early stage of T2D (Merdzo *et al*., 2017). The lower VDAC in the cerebral microvessels of the ZDFO rats might represent a compensatory mechanism to minimize the detrimental consequences of increased oxidative stress in T2D.

Inhibition of VDAC in diabetes treatment

Inhibition of VDAC and VDAC-mediated apoptosis offers a potential strategy for combating diseases like diabetes. There are many known inhibitors of VDAC which affects its voltage gating (Holmuhamedov and Lemasters, 2009; Ben-Hail *et al.*, 2016), however, still few experimental data are available with respect to their use for diabetes treatment. Metformin, an antidiabetic drug has been shown to counteract VDAC1 induction by direct blocking it in db/db mice and restored the impaired ATP generation along with glucose-stimulated insulin secretion in T2D islets (Zhang *et al.* 2019a). In the study by Zhang *et al.* (2019a), two other VDAC inhibitors, VBIT-4 and AKOS (AKOS022075291) were also studied. VBIT-4 and AKOS are novel group of piperarazine- and piperidine-based compounds which directly interact with VDAC1 thereby inhibiting VDAC activities that are associated with metabolite transport, oligomerization as well as activities related to changes in intracellular calcium levels, reactive oxygen species and mitochondrial membrane potential due to mitochondria dysfunction. Recent study by Pittala *et al.* (2020) demonstrated that treatment of T2D model with VDAC1-based peptide, R-Tf-D-LP4 restored the elevated blood glucose levels and caused an increase in number and average size of islets and their insulin content suggesting that this peptide has potential for diabetes treatment. Patent filed by the inventors (Salehi *et al.*, 2018) on 2017-12-21, disclosed the use of substituted piprazine and piperidine derivatives as specific inhibitors of VDAC1 for preventing the progression of and treating prediabetes

and diabetes.

Apart from chemical compounds, natural plant products are another alternative for treating diabetes and diabetic complications without adverse side effects. This alternative has been one of the areas of interest for targeting VDAC in the treatment of diabetes and other diseases. Curcumin, an active compound found in turmeric, has glucose-lowering effect and improve β-cell function in T2D (Wickenberg *et al.,* 2010). This promising antidiabetic property has been postulated to be due to the ability to bind to and inhibit VDAC (De Marchi *et al.,* 2020). Further, methyl jasmonate, a cyclopentanone lipid-belonging to the family of plant stress hormones, can detach hexokinase from VDAC1 from mitochondria causing a dissociated glycolysis, decrease in ATP and release of cytochrome *c* ultimately leading to cell death (Goldin *et al.,* 2007; 2008). Therefore, targeting VDAC particularly in the β-cell, can prove beneficial in restoring normal β-cell functions in diabetes.

Conclusion

Diabetes is associated with enhanced apoptotic cell death which has been linked to a spectrum of diabetic complications. VDAC a key mitochondrial membrane protein involved in apoptosis has been explored for its contribution to the pathogenesis of diabetes. Several studies have demonstrated that inhibition of VDAC prevents progression of diabetes. Thus, targeting VDAC may provide a novel therapeutic strategy for the treatment of diabetes and its complications.

Conflict of interest

The authors claim that there is no conflict of interest.

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References

Abu-Hamad, S., Arbel, N., Calo, D., Arzoine L., Israelson, A., Keinan, N., Ben-Romano, R., Friedman, O. and Shoshan-Barmatz, V. 2009. The VDAC1 N-terminus is Essential Both for Apoptosis and the Protective Effect of Anti-apoptotic Proteins. *Journal of Cell Science,* 122: 1906-1916.

Ahmed, M., Muhammed, S.J., Kessler, B. and Salehi, A. 2010. Mitochondrial Proteome Analysis Reveals Altered Expression of Voltage Dependent Anion Channels in Pancreatic β-cells Exposed to High Glucose. *Islets*, 2(5): 283-292.

Allen, D.A., Yaqoob, M.M. and Harwood, S.M. 2005. Mechanisms of High-glucose-induced Apoptosis and its Relationship to Diabetic Complications. *The Journal of Nutritional Biochemistry,* 16(12): 705-713.

American Diabetes Association. 2009. Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care*, 32(Suppl 1): S62-S67.

Anflous-Pharayra, K., Lee, N., Armstrong, D.L. and Craigen, W.J. 2011. VDAC3 has Differing Mitochondrial Functions in Two Types of Striated Muscles. *Biochimica et Biophysica Acta,* 1807: 150-156.

Arbel, N., Ben-Hail, D. and Shoshan-Barmatz, V. 2012. Mediation of the Antiapoptotic Activity of Bcl-xL Protein upon Interaction with VDAC1 Protein. *Journal of Biological Chemistry,* 287: 23152-23161.

Arzoine, L., Zilberberg, N., Ben-Romano, R. and Shoshan-Barmatz, V. 2008. Voltage-Dependent Anion Channel 1-based Peptides Interact with Hexokinase to Prevent Its Anti-apoptotic Activity. *Journal of Biological Chemistry,* 284: 3946-3955.

Barber, A.J., Gardner, T.W. and Abcouwer, S.F. 2011. The Significance of Vascular and Neural Apoptosis to the Pathology of Diabetic Retinopathy. *Investigative Ophthalmology & Visual Science,* 52(2): 1156-1163.

Ben-Hail, D., Begas-Shvartz, R., Shalev, M., Shteinfer-Kuzmine, A., Gruzman, A., Reina, S., De Pinto, V. and Shoshan-Barmatz, V. 2016. Novel Compounds Targeting the Mitochondrial Protein VDAC1 Inhibit Apoptosis and Protect Against Mitochondria Dysfunction. *Journal of Biological Chemistry,* 291: 24986-25003.

Bergmann, A. 2002. Survival Signaling Goes BAD. *Developmental Cell*, 3(5): 607-608.

Caterino, M., Ruoppolo, M., Mandola, A., Costanzo, M., Orru, S. and Imperlini, E. 2017. Protein–Protein Interaction Networks as a New Perspective to Evaluate Distinct Functional Roles of Voltage-Dependent Anion Channel Isoforms. *Molecular BioSystems,* 13: 2466-2476.

Cnop, M., Welsh, N., Jonas, J.C., Jo¨rns, A., Lenzen, S. and Eizirik, D.L. 2005. Mechanisms of Pancreatic Beta-Cell Death in Type 1 and Type 2 Diabetes: Many Differences, Few Similarities. *Diabetes*, 54: S97-S107.

Colombini, M. 2004. VDAC: The Channel at the Interface Between Mitochondria and the Cytosol. *Molecular & Cellular Biochemistry*, 256 : 107-115.

Danial, N.N., Gramm, C.F., Scorrano, L., Zhang, C.Y., Krauss, S., Ranger, A.M., Datta, S.R., Greenberg, M.E., Licklider, L.J., Lowell, B.B., Gygi, S.P. and Korsmeyer, S.J. 2003. BAD and Glucokinase Reside in a Mitochondrial Complex that Integrates Glycolysis and Apoptosis. *Nature*, 424: 952-956.

De Marchi, U., Fernandez-Martinez, S., de la Fuente, S., Wiederkehr, A. and Santo-Domingo, J. 2020. Mitochondrial Ion Channels in Pancreatic Beta-cells: Novel Pharmacological Targets for the Treatment of Type 2 Diabetes. *Brazilian Journal of Pharmacology*, doi:10.1111/bph.15018 (2020) (Epub ahead of print).

De Pinto, V., Guarino, F., Guarnera, A., Messina, A., Reina, S., Tomasello, F.M., Palermo, V., Mazzoni, C. 2010. Characterization of Human VDAC Isoforms: A Peculiar

Function for VDAC3?. *Biochimica et Biophysica Acta*, 1797(6-7): 1268-1275.

De Pinto, V., Reina, S., Gupta, A., Messina, A. and Mahalakshmi, R. 2016. Role of Cysteines in Mammalian VDAC Isoforms' Function. *Biochimica et Biophysica Acta,* 1857: 1219-1227.

De Stefani, D., Bononi, A., Romagnoli, A., Messina, A., De Pinto, V., Pinton, P. and Rizzuto R. 2012. VDAC1 selectively Transfers Apoptotic $Ca^{(2+)}$ Signals to Mitochondria. *Cell Death Differentiation*, 19(2): 267-273.

Feenstra, D.J., Yego, E.C. and Mohr, S. 2013. Modes of Retinal Death in Diabetic Retinopathy. *Journal of Clinical & Experimental Ophthalmology*, 4(5): 298.

Forbes, J.M. and Cooper, M.E. 2013. Mechanism of Diabetic Complications. *Physiological Reviews,* 93(1): 137-188.

Goldin, N., Arzoine, L., Heyfets, A., Israelson, A., Zaslavsky, Z., Bravman, T., Bronner, V., Notcovich, A., Shoshan-Barmatz, V. and Flescher, E. 2008. Methyl Jasmonate Binds to and Detaches Mitochondria-bound Hexokinase. *Oncogene*, 27(34): 4636-4643.

Goldin, N., Heyfets, A., Reischer, D. and Flescher, E. 2007. Mitochondria-mediated ATP Depletion by Anti-cancer Agents of the Jasmonate Family. *Journal of Bioenergetics & Biomembranes,* 39: 51-57.

Gong, D., Chen, X., Middleditch, M., Huang, L., Vazhoor Amarsingh, G., Reddy, S., Lu, J., Zhang, S., Ruggiero, K., Phillips, A.R. and Cooper, G.J. 2009. Quantitative Proteomic Profiling Identifies New Renal Targets of Copper (II)-selective Chelation in the Reversal of Diabetic Nephropathy in Rats. *Proteomics*, 9: 4309-4320.

Guarino, F., Specchia, V., Zapparoli, G., Messina, A., Aiello, R., Bozzetti, M.P. and De Pinto, V. 2006. Expression and Localization in Spermatozoa of the Mitochondrial Porin Isoform 2 in *Drosophila melanogaster*. *Biochemical & Biophysical Research Communications*, 346: 665-670.

Habib, S.L. 2013. Diabetes and Renal Tubular Cell Apoptosis. *World Journal of Diabetes*, 4(2): 27-30.

Hinsch, K.D., De Pinto, V., Aires, V.A., Schneider, X., Messina, A. and Hinsch, E. 2004. Voltage-dependent Anion-selective Channels VDAC2 and VDAC3 are Abundant Proteins in Bovine Outer Dense Fibers, a Cytoskeletal Component of the Sperm Flagellum. *Journal of Biological Chemistry*, 79: 15281-15288.

Holmuhamedov, E. and Lemasters, J.J. 2009. Ethanol Exposure Decreases Mitochondrial Outer Membrane Permeability in Cultured Rat Hepatocytes. *Archives of Biochemistry & Biophysics*, 481: 226-233.

Jones, S.P., Zachara, N.E., Ngoh, G.A., Hill, B.G., Teshima, Y., Bhatnagar, A., Hart, G.W. and Marban, E. 2008. Cardioprotection by N-acetylglucosamine linkage to Cellular Proteins. *Circulation*, 117: 1172-1182.

Kang, B.P., Urbonas, A., Baddoo, A., Baskin, S., Malhotra, A. and Meggs, L.G. 2003. IGF-1 Inhibits the Mitochondrial Apoptosis Program in Mesangial Cells Exposed to High Glucose. *American Journal of Physiology- Renal Physiology*, 285: F1013-F1024.

Kim, J., Gupta, R., Blanco, L.P., Yang, S., Shteinfer-Kuzmine, A., Wang, K., Zhu, J., Yoon, H.E., Wang, X., Kerkhofs, M., Kang, H., Brown, A.L., Park, S., Xu, X., van Rilland, E.Z., Kim, M.K., Cohen, J.I., Kaplan, M.J., Shoshan-Barmatz, V. and Chung, J.H. 2019. VDAC Oligomers Form Mitochondrial Pores to Release mtDNA Fragments and Promote Lupus-like Disease. *Science*, 366: 1531-1536.

Kim, W.H., Lee, J.W., Suh, Y.H., Hong, S.H., Choi, J.S., Lim, J.H., Song, J.H., Gao, B. and Jung, M.H. 2005. Exposure to chronic high glucose induces β-cell apoptosis through decreased interaction of glucokinase with mitochondria. *Diabetes*, 54(9): 2602-2611.

Kroemer, G., Galluzzi, L. and Brenner, C. 2007. Mitochondrial Membrane Permeabilization in Cell Death. *Physiological Reviews,* 87:99-163.

Lee, S.C. and Pervaiz, S. 2007. Apoptosis in the Pathophysiology of Diabetes Mellitus. *International Journal of Biochemistry & Cell Biology*, 39(3): 497-504.

Leist, M. and Jaattela, M. 2001. Four Deaths and a Funeral: from Caspases to Alternative Mechanism. *Nature Reviews Molecular Cell Biology,* 2: 589-598.

Li, X., Wang, H., Yao, B., Xu, W., Chen, J. and Zhou, X. 2016. lncRNA H19/miR-675 axis Regulates Cardiomyocyte Apoptosis by Targeting VDAC1 in Diabetic Cardiomyopathy. *Scientific Reports*, 6: 36340.

Majewski, N., Nogueira, V., Robey, R.B. and Hay, N. 2004. Akt Inhibited Apoptosis Downstream of BID Cleavage *via* a Glucose-dependent Mechanism Involving Mitochondrial Hexokinases. *Molecular & Cellular Biology*, 24: 730-740.

Majumder, S., Slabodnick, M., Pike, A., Marquardt, J. and Fisk, H.A. 2012. VDAC3 Regulates Centriole Assembly by Targeting Mps1 to Centrosomes. *Cell Cycle*, 11: 3666 -3678.

McKenzie, M.D., Jamieson, E., Jansen, E.S., Scott, C.L., Huang, D.C., Bouillet, P., Allison, J., Kay, T.W., Strasser, A. and Thomas, H.E. 2010. Glucose Induces Pancreatic Islet Cell Apoptosis that Requires the BH3-only Proteins bim and puma and Multi-BH Domain Protein Bax. *Diabetes*, 59: 644-652.

Merdzo, I., Rutkai, I., Sure, V.N., McNulty, C.A., Katakam, P.V. and Busij, D.W. 2017. Imaired Mitochondrial Respiration in Large Cerebral Arteries of Rats with Type 2 Diabetes. *Journal of Vascular Research*, 54(1): 1-12.

Mishra, R., Emancipator, S.N., Kern, T. and Simonson, M.S. 2005. High Glucose Evokes an Intrinsic Proapoptotic Signaling Pathway in Mesangial Cells. Kidney International, 67(1): 82-93.

Mostyn, A., Pearce, S., Stephenson, T. and Symonds, M.E. 2004. Hormonal and Nutritional

Regulation of Adipose Tissue Mitochondrial Development and Function in the Newborn. Experimental and Clinical Endocrinology and Diabetes, 112: 2-9.

Nordlie, R.C., Foster, J.D. and Lange, A.J. 1999. Regulation of Glucose Production by the Liver. *Annual Reviews in Nutrition*, 19: 379-406.

Ouyang, C., You, J. and Xie, Z. 2014. The Interplay Between Autophagy and Apoptosis in the Diabetic Heart. *Journal of Molecular and Cellular Cardiology*, 71:71-80.

Peng, C., Ma, J., Gao, X., Tian, P., Li, W. and Zhang, L. 2013. High Glucose Induced Oxidative Stress and Apoptosis in Cardiac Microvascular Endothelial Cells Are Regulated by FoxO3a. *PLoS ONE*, 8(11): e79739.

Pittala, S., Levy, I., De, S., Kumar Pandey, S., Melnikov, N., Hyman, T., & Shoshan-Barmatz, V. 2020. The VDAC1-based R-Tf-D-LP4 Peptide as a Potential Treatment for Diabetes Mellitus. *Cells*, 9(2): 481.

Reina, S., Palermo, V., Guarnera, A., Guarino, F., Messina, A., Mazzoni, C. and De Pinto, V. 2010. Swapping of the N-terminus of VDAC1 with VDAC3 restores Full Activity of the Channel and Confers Anti-aging Features to the Cell. *FEBS Letters*, 584: 2837-2844.

Salehi, A. 2010. Mitochondrial Proteome Analysis Reveals Altered Expression of Voltage Dependent Anion Channels in Pancreatic Beta-cells Exposed to High Glucose. *Islets*, 2: 283-292.

Salehi, A., Wollheim C.B. and Shoshan-Barmatz, V. 2018. The National Institute for Biotechnology in the Negev Ltd. PCT/IL2017/051379 filed December 21, 2017 and issued June 28, 2018.

Sampson, M.J., Decker, W.K., Beaudet, A.L., Ruitenbeek, W., Armstrong, D., Hicks, M.J. and Craigen, W.J. 2001. Immotile Sperm and Infertility in Mice Lacking Mitochondrial Voltage-dependent Anion Channel Type 3. *Journal of Biological Chemistry*, 276: 39206-39212.

Sasaki, K., Donthamsetty, R., Heldak, M., Cho, Y.E., Scott, B.T. and Makino, A. 2012. VDAC: Old Protein with New Roles in Diabetes. *American Journal of Physiology Cell Physiology,* 303 (10): C1055-C1060.

Schattenberg, J.M. and Schuchmann, M. 2009. Diabetes and Apoptosis. *Liver*, 14: 1459.

Sepassi, L.,Sasaki, K., Heldak, M. and Makino, A. 2013. Role of VDAC in Coronary Endothelial Cell Dysfunction in Type 1 Diabetic Mice. *The FASEB Journal*, 27: 913.2-913.2.

Shoshan-Barmatz, V., De Pinto, V., Zweckstetter, M., Raviv, Z., Keinan, N. and Arbel, N. 2010. VDAC, a Multi-functional Mtochondrial Protein Regulating Cell Life and Death. *Molecular Aspects of Medicine*, 31(3): 227-285.

Shoshan-Barmatz, V., Kmita, H. and Lemasters, J.J. 2017. Editorial: Uncovering

the Function of the Mitochondrial Protein VDAC in Health and Disease: From Structure- Function to Novel Therapeutic Strategies. *Frontiers in Oncology*, 7: 320.

Shoshan-Barmatz, V., Krelin, Y. and Shteinfer-Kuzmine, A. 2018. VDAC1 Functions in Ca2+ Homeostasis and Cell Life and Death in Health and Disease. *Cell Calcium*, 69: 81-100.

Tajeddine, N., Galluzzi, L., Kepp, O., Hangen, E., Morselli, E., Senovilla, L., Araujo, N., Pinna, G., Larochette, N., Zamzami, N., Modjtahedi, N., Harel-Bellan, A. and Kroemer, G. 2008. Hierarchical Involvement of Bak, VDAC1 and Bax in Cisplatin-induced Cell Death. *Oncogene*, 27: 4221-4232

Teplova, V.V., Odinokova, I.V. and Holmuhamedov, E.L. 2011. Isoforms of Voltage-dependent Anion Channel of the Outer Mitochondrial Membrane and Experimental Models to Study Their Physiological Role. *Biochemistry (Moscow) Supplement Series A: Membrane and Cell Biology*, 5(2): 111-120.

Turko, I.V. and Murad, F. 2003. Quantitative Protein Profiling in Heart Mitochondria from Diabetic Rats. *Journal of Biological Chemistry*, 278: 35844-35849.

Ujwal, R., Cascio, D., Colletier, J.P., Faham, S., Zhang, J., Toro, L., Ping, P. and Abramson, J. 2008. The Crystal Structure of Mouse VDAC1 at 2.3 A Resolution Reveals Mechanistic Insights into Metabolite Gating. *Proceedings of the National Academy of Sciences USA*, 105: 17742-17747.

Vander Heiden, M.G., Chandel, N.S., Li, X.X., Schumacker, P.T., Colombini, M. and Thompson C.B. 2000. Outer Mitochondrial Membrane Permeability Can Regulate Coupled Respiration and Cell Survival. *Proceedings of the National Academy of Sciences USA*, 97: 4666-4671.

Wali, J.A., Masters, S.L. and Thomas, H.E. 2013. Linking Metabolic Abnormalities to Apoptotic Pathway in beta Cells in Type 2 Diabetes. *Cells*, 2(2): 266-283.

Wang, L., Wu, C. G., Fang, C. Q., Gao, J., Liu, Y. Z., Chen, Y., Chen, Y. N., and Xu, Z. G. 2013. The protective effect of α-Lipoic acid on mitochondria in the kidney of diabetic rats. *International Journal of Clinical and Experimental Medicine*, 6(2): 90-97.

Wickenberg, J., Ingemansson, S.L. and Hlebowicz, J. 2010. Effects of *Curcuma longa* (turmeric) on Postprandial Plasma Glucose and Insulin in Healthy Subjects. *Nutrition Journal*, 9: 43.

Yuan, S., Fu, Y., Wang, X., Shi, H., Huang, Y., Song, X., Li, L., Song, N. and Luo, Y. 2008. Voltage-Dependent Anion Channel 1 is Involved in Endostatin-induced Endothelial Cell Apoptosis. *The FASEB Journal*, 22: 2809-2820.

Zhang, E., Mohammed Al-Amily, I., Mohammed, S., Luan, C., Asplund, O., Ahmed, M., Ye, Y., Ben-Hail, D., Soni, A., Vishnu, N., Bompada, P., De Marinis, Y., Groop, L., Shoshan-Barmatz, V., Renström, E., Wollheim, C. B., and Salehi, A. 2019a.

Preserving Insulin Secretion in Diabetes by Inhibiting VDAC1 Overexpression and Surface Translocation in β Cells. *Cell Metabolism*, 29(1): 64-77.e6.

Zhang, J., Guo, Y., Ge, W., Zhou, X. and Pan, M. 2019b. High Glucose Induces Apoptosis of HUVECs in a Mitochondriadependent Manner by Suppressing Hexokinase 2 Expression. *Experimental and Therapeutic Medicine*, 18: 621-629.

Zhaoyu, J. and El-Deiry, W.S. 2005. Overview of Cell Death Signalling Pathways. *Cancer Biology & Therapy*, 4(2): 147-171.