

Role of Haptoglobin in Health and Disease: A Focus on Diabetes

Mark MacKellar¹ and David J. Vigerust^{1,2}

■ **IN BRIEF** Prospective identification of individuals with diabetes who are at greatest risk for developing complications would have considerable public health importance by allowing appropriate resources to be focused on those who would benefit most from aggressive intervention. Haptoglobin (Hp) is an acute-phase protein that is crucial for the elimination of free hemoglobin and the neutralization of oxidative damage. In the past two decades, associations have been made between polymorphisms in Hp and complications arising from diabetes. Individuals with polymorphism in Hp have been shown to have significantly higher risk of developing cardiovascular disease. This review summarizes the current literature on the role of Hp in health and disease, with a focus on diabetes.

Recent epidemiological studies have showed that the diabetes epidemic affects 415 million people worldwide, and this figure is expected to increase to nearly 642 million people by 2040 (1). Patients afflicted with diabetes often are more susceptible to a host of multi-organ complications that arise as a result of microvascular and macrovascular dysfunction. These complications equate to the leading cause of morbidity and mortality, in the form of accelerated atherosclerotic disease. Dysregulation in glycemic control and subsequent alterations to the vascular endothelium are known to be associated with all grades of atherosclerotic disease (2,3). Accelerated atherosclerosis is prevalent in this patient population, contributing to >75% of the mortality seen among people with diabetes (4).

In the past decade, interventions to obtain stricter glycemic control have been attempted with debatable success. In addition to stricter glycemic control measures, attempts have been made to reduce cardiovascular risk by implementing lifestyle mod-

ifications and medication regimens to reduce hyperlipidemia and hypertension. One area of considerable interest in the clinical and research community is the development of improved screening tools to identify and mitigate cardiovascular risk. One such area that has had considerable interest is the predictive value of haptoglobin (Hp) genotype screening in cardiovascular risk.

Polonovski and Jayle first described the biochemical properties of Hp more than 75 years ago (5–7). Since that initial description, numerous important biological functions have been described for Hp. Hp is an acute-phase α_2 -glycoprotein with a major biological function of binding free hemoglobin (Hb) with very high affinity to prevent the loss of iron following intravascular hemolysis (8,9) and to prevent Hb-mediated renal injury (10–12). The binding of Hp to free Hb forms a stable complex with very high affinity that is cleared from circulation by the reticuloocyte system and CD163-positive macrophages, Kupffer cells, and hepatocytes (13,14).

¹MyGenetx Clinical Laboratories, Franklin, TN

²Vanderbilt University School of Medicine, Department of Neurological Surgery, Nashville, TN

Corresponding author: David J. Vigerust, dvigerust@mygenetx.com

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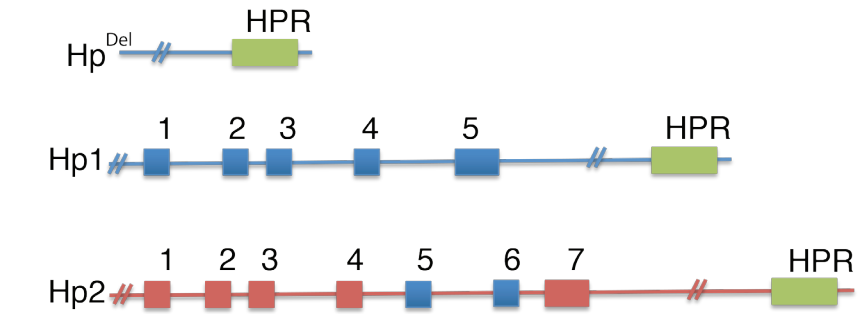
In its role as a clearance protein, Hp removes the oxidative potential of the iron contained in the Hb molecule.

Hp is present in the serum of all mammals, but polymorphism is found only in humans (15). The allelic differences seen in humans arise from a crossover duplication of exons 3 and 4, resulting in an Hp1 allele with 5 exons and an Hp2 allele with 7 exons (Figure 1) (16). Three major genotypes are produced and have been identified by gel electrophoresis: Hp1-1, Hp1-2, and Hp2-2 (17,18). Hp^{Del} has also been described in Japanese, Chinese, and Korean populations but has not been found in African or European populations (19–21). Several minor genotypes (Hp1-Johnson, Hp1-M, and Hp0) have also been described (22). The three predominant genotypes display differing structures and biological effects, as described below.

Molecular Structure and Regulation

The human gene for Hp is located on chromosome 16q22 and consists of three structural alleles, the products of which are Hp1F, Hp1S, and Hp2 (23). The products of the Hp1F and Hp1S alleles differ by only one amino acid, whereas the Hp2 allele is the result of a fusion of the Hp1F and Hp1S alleles. Transcription and translation occurs as a single polypeptide that is post-translationally processed into a smaller α chain and a larger β chain that are linked by a disulfide bridge (24). However, the presence of the Hp1 and Hp2 alleles in humans gives rise to Hp1-1 dimers, Hp1-2 heterodimers, and Hp2-2 dimers. The estimated frequency of Hp1-1 is 15–18%, Hp2-1 is 46%, and Hp2-2 is 38% (25).

Hp synthesis is principally conducted in the liver, but expression is also seen in the lung, skin, spleen, kidney, and adipose tissue (26–28). The expression of Hp can be increased in the presence of growth hormone, insulin, and bacterial endotoxin and in the expression of macrophage-



■ **FIGURE 1.** Structural representation of the Hp alleles Del, 1, and 2. Hp exonic sequences are denoted by numbered and shaded boxes. Intronic sequences are denoted by a solid line. Exons 3 and 4 of the Hp1 allele have been duplicated in the Hp2 allele, giving rise to exons 3–6.

produced proinflammatory cytokines such as interleukin (IL)-1, IL-6, and tumor necrosis factor- α (TNF- α) (14,29,30). In addition to induction by cytokines, glucocorticoids, catecholamines, and hypoxia also can activate the expression of Hp (31,32). Despite largely being a serum protein, Hp is also detected in urine, synovial fluid, ascites fluid, cerebrospinal fluid, and pleural fluid (33).

Functions of Hp

The hallmark function of Hp is to facilitate Hb clearance. Hb is the prominent blood protein involved in transporting oxygen in the circulatory system and mediates reactive oxygen and nitrogen species detoxification (34,35). Free Hb released into the blood is a natural phenomenon associated with intravascular hemolysis that occurs during the destruction of senescent red blood cells (RBCs), which occurs at a rate of 2×10^6 RBCs per second (36,37). A variety of severe complications can result from intravascular hemolysis when accompanied by other comorbidities such as diabetes, infectious disease, trauma, and cancer (38,39). Hp forms a strong noncovalent complex with Hb (Hp-Hb complex) that results in its removal via the reticuloendothelial system and receptor-mediated endocytosis via CD163 on hepatocytes, Kupffer cells, and tissue macrophages (40–43).

There is sufficient Hp in circulation (38–208 mg/dL) to bind and clear 3 g of Hb, which would prevent free Hb circulation in the body (44). CD163 expression by these cells is induced by inflammation and the release of cytokines such as IL-1 and IL-6. IL-6 plays an especially important function in that it is important in the stimulation of Hp production and the modulation of CD163 expression on the cell surface (45–47). CD163 is downregulated by TNF- α , IL-4, and interferon γ (48). Regulation of this process is tightly controlled by the release of immunomodulatory molecules.

Hb that is bound with Hp for clearance is internalized, processed, and degraded to release heme for further processing by hemoxygenase-1 to release iron where it can be recycled to construct new Hb proteins. When not bound to Hb, Hp is cleared from the plasma in 3–5 days. When bound to Hb, the average time for removal of the complex is ~20 minutes (49,50). Hp is not recycled during this clearance process, but rather is degraded, and de novo protein production occurs to resupply the blood and tissue (51). In the absence of a clearance mechanism, free Hb can catalyze the formation of free radicals and mediate the destruction of cellular constituents and extracellular macromolecules and promote the oxidation of LDL cholesterol (37).

Antioxidant Activity

Hp binds to free Hb with perhaps the highest affinity in nature (9,52,53). A variety of unfavorable consequences can arise when free iron is present in the circulation and in tissues. For some time, it has been known that iron overload can contribute to the development of diabetes and atherosclerosis (54–57). The pathophysiology with respect to iron arises from the ability of iron to participate in the generation of powerful oxidant species such as hydroxyl radical (58). Iron participates in the Haber-Weiss reaction to convert reactive oxygen species (ROS) such as superoxide and hydrogen peroxide to more powerful species such as hydroxyl radical (56). Free iron can be a detrimental catalyst in lipid peroxidation because it can both initiate and amplify the process of lipid peroxidation.

The initiation step can be induced by two different iron-dependent mechanisms. The hydroxyl radical-dependent mechanism, which has been adopted by most as the predominant mechanism for lipid peroxidation (59), and an alternate hydroxyl radical-independent mechanism, which has also been proposed and places iron-oxygen complexes rather than hydroxyl radical as the initiator of lipid peroxidation (60). These conversions can have damaging effects to the tissue.

The ability of Hp to reduce the tissue-damaging effects of free radicals is genotype-dependent. Investigations by Melamed-Frank et al. demonstrate that Hp1-1 has a greater capacity to protect against oxidative damage despite the fact that all three genotypes have the same binding affinity (61). The crucial difference is the accessibility of the Hp molecule to the extravascular space. Size differences in the molecules serve as an exclusionary mechanism; consequently, in individuals with Hp2-1 or Hp2-2, free Hb remains in circulation for a protracted period of time and causes greater oxidative stress (61). The antioxidant capacity of Hp

in people with diabetes is of special concern. In particular, the Hp2-2 genotype is considered to be a major susceptibility gene for people with type 2 diabetes because it results in reduced antioxidant capacity (62).

Immunoregulatory Activity

The role of Hp as an acute-phase protein brings to light its possible immunoregulatory activity. Dating back as far as 1968 with the work of Nevo and Stutton (63) and, more recently, the work of Langlois and Delanghe (64), individuals of Hp2-2 genotype demonstrated enhanced immune function. For example, Hp2-2 individuals display a greater response to vaccination by producing higher levels of protective antibodies (65). Hp also has been described by Langlois and Delanghe as having inhibitory activity in the synthesis of prostaglandins and, as a result, having anti-inflammatory properties (64).

Hp has been shown to have powerful regulatory activity on lymphocytes. For example, Arredouani et al. (66) and Guetta et al. (67) have demonstrated that Hp plays a role in the balance between T helper-1 (Th1) and T helper-2 (Th2) by heavily promoting a strong Th1 cell response. These studies demonstrated that the Hp1 allele, when complexed to Hb, stimulates the secretion of significantly more IL-6 and IL-10 than the Hp2 allele. A Th1 response is more effective in protecting against intracellular parasites and inhibits the release of Th2 cytokines, which are responsible for defending against extracellular pathogens. These findings suggest that patients with Hp1 alleles are more efficient at protecting against infection.

In the context of immune-mediated diseases, Crohn's disease, ulcerative colitis, rheumatoid arthritis, and systemic lupus erythematosus all have been linked with polymorphisms in Hp. For example, pro-haptoglobin-2 (proHp), which is proteolytically cleaved to Hp2 by site-specific cleavage, has been asso-

ciated with celiac disease and type 1 diabetes (68,69). Recently, studies by Zhang et al. (70) and Moreno-Navarrete et al. (71) have suggested that serum levels of proHp are elevated in people with type 2 diabetes and that circulating levels of proHp contribute to insulin resistance in obesity. The elevations in proHp are associated with intestinal permeability, dyslipidemia, inflammation, and insulin resistance.

Angiogenesis and Lymphangiogenesis

Impaired wound healing is a known major complication of diabetes and is caused by apoptosis, cellular infiltration, and reduced angiogenesis. Recent studies have demonstrated reduced lymphangiogenesis and angiogenesis during diabetic wound healing (72–74). Hp is known to play a role in angiogenesis according to Cockerill et al. (75). A study by Oh et al. (76) demonstrated that proHp can upregulate the expression of vascular endothelial growth factor (VEGF) and VEGF receptor 2 and increase endothelial sprouting and branching. These findings suggest that proHp can promote angiogenesis via the VEGF signaling pathway.

Cid et al. (77) observed that sera from patients with systemic vasculitis stimulated angiogenesis in an *in vitro* model. Serum Hp level in vasculitis patients was shown to correlate with both disease and angiogenic activity. The increased levels of Hp found in chronic inflammatory conditions may play an important role in tissue repair. In systemic vasculitis, for example, Hp might compensate for ischemia by promoting the development of collateral vessels (77).

Neovascularization resulting from angiogenesis may be pathologically important in the context of diabetes. Pathological angiogenesis enhances disease progression and increases macrophage infiltration and vessel wall thickness, leading to hypoxia and interplaque rupture (78,79). Lipid-rich RBC membranes and free

Hb are physiologically detrimental in the context of diabetes and atherosclerosis. Neovascularization of the atherosclerotic plaque may be driven by Hp or proHp, leading to greater risk in people with diabetes.

Blockade of Nitric Oxide

Nitric oxide (NO) is a gas produced by a variety of cell types that is involved in several important physiological activities. NO is involved in vascular tone, modulates neurotransmitter function in the central and peripheral nervous systems, and participates in cellular defense and platelet aggregation (80,81). Both free Hb and the Hp-Hb complex inactivate NO, whereas Hp does not affect NO. As a result, an increase in the level of circulating Hb or Hp-Hb complex may result in the depletion of NO and the lack of endothelial relaxation contributing to cardiovascular disease (CVD).

Patients with an Hp1-1 genotype may experience benefits over Hp2-1 or Hp2-2 carriers because the Hp1-1 Hp-Hb complex will be cleared from circulation more efficiently than other Hp complexes. The more rapid clearance enhances the availability of NO to carry out its physiological functions. One illustration of the benefits of one Hp genotype over another was seen in a recent report by Sertorio et al. (82) showing that, in patients with preeclampsia, Hp1-1 played a protective role by reducing NO scavenging. Patients with Hp2-1 and Hp2-2 appeared to have aggravation of preeclampsia, in part because of reduced NO bioavailability.

Disease Implications of Hp Polymorphism

Infectious Disease

Diabetes is a predisposing factor for infections. People with diabetes have a two to four times greater risk of systemic infection than people without diabetes (83,84). Variation in Hp genotype is also implicated as a contribution to mortality from both extracellular and intracellular pathogens. For example, several studies

have demonstrated that patients can have a greater susceptibility to malaria and the development of severe complications depending on their Hp genotype (85–88).

Pathogens such as *Corynebacterium diphtheriae* and *Staphylococcus aureus*, which require iron for growth, are known to take advantage of the Hp-Hb complex for their metabolic activity (89,90). The reduced activity of Hp2 to clear free Hb may allow these bacteria to use this iron source, contributing to enhanced colonization and growth. Pishchany et al. (91) have demonstrated that *S. aureus* can grow in a manner that is entirely dependent on the ability to bind Hb, extract heme, pass heme through the bacterial cell envelope, and degrade heme in the cytoplasm. The ability of different bacteria to use iron from Hb may allow for the generation of soft tissue infection when free heme is not removed after RBC hemolysis.

People with diabetes are especially susceptible to bacterial infections, especially urinary tract, respiratory, and soft tissue infections. For example, *S. aureus* is known to cause both respiratory infections and urinary tract infections that can lead to bacterial nephritis. Additionally, *S. aureus* is the most common soft tissue infection in people with diabetes (83,92,93). These findings could be especially important in the context of diabetes complications, CVD, surgical interventions, and trauma. In general, host innate immune mechanisms, including Hp, would restrict the accessibility of iron to pathogens, thereby removing one important bacterial growth requirement (94,95).

Finally, Hp2-2 patients also have been found to have higher mortality and poorer outcomes from tuberculosis and Hansen's disease (leprosy) than other genotypes (88,95–97). Given its role as an acute-phase protein, the participation of Hp in various pathogenic infections is well documented, and the specific genotype that a patient carries has direct

bearing on the ultimate outcome of infection.

Hp genotype has also been associated with intracellular pathogens. Diabetes has been recognized as an important risk factor for several intracellular pathogens (98). For example, patients with Hp2-2 have a higher degree of mortality from HIV, with a median survival time of 11 years for people with Hp1-1 or Hp2-1 compared to 7.3 years for those with Hp2-2. Hp2-2 patients also had significantly higher HIV viral titers than those with other genotypes (99). Hp genotype also has been investigated in HIV patients in the development of Kaposi sarcoma (KS). KS is caused by herpes virus 8, an opportunistic infection rarely seen outside of immunocompromised patients. Speeckaert et al. (100) examined the effect of Hp genotype and demonstrated that the Hp1-1 genotype conferred the greatest risk to the development of KS, followed by Hp2-2 and Hp2-1. In contrast, Speeckaert et al. (101) in an earlier study demonstrated that Hp1-1 provided the greatest protection from Epstein-Barr virus, and Hp2-2 has the greatest association as determined by serum titer. In animal models of influenza, there is significant evidence that Hp is upregulated in clinical disease. This has been found in pigs and ferrets, which share a lung physiology similar to that of humans (102,103). Significant observations regarding bovine respiratory disease have been associated with increased Hp production (104). These observations in respiratory infection are especially relevant given the recent report of Hp as part of the human lung surfactant system, in which it co-localizes with surfactant protein B (105). As seen through these few examples, susceptibility to infectious disease is, in part, dependent on Hp genotype, and this is especially important in people with diabetes and Hp polymorphism.

Neurology

Diabetes has been associated with dementia and neurodegenerative

disorders such as Alzheimer's disease (AD) and Parkinson's disease (PD) (106,107). Although the mechanisms remain unclear, type 2 diabetes can exacerbate and lead to progression of the neurodegenerative process. One possible common mechanism is oxidative stress (108). In addition to this commonality in pathogenesis, there is also evidence that AD itself can promote insulin resistance within the brain (108).

Given the common thread of oxidative stress and the role of Hp in the management of oxidative reactions, several recent studies have demonstrated the relationship of Hp with neurodegenerative pathologies. In fact, Hp genotype has been implicated in greater susceptibility to idiopathic PD (109,110). There are also several reports of Hp involvement in the pathogenesis of AD. The accumulation of β -amyloid in the brain is a driving force for AD pathogenesis. Spagnuolo et al. previously reported that Hp could bind to apolipoprotein E (ApoE) and impair its function in cholesterol homeostasis (111). Immunoprecipitation and immunoblotting has shown that Hp and β -amyloid form complexes in brain tissue from patients with AD. The interaction between ApoE and β -amyloid was shown to be crucial for limiting β -amyloid neurotoxicity and for promoting its clearance. Spagnuolo et al. further demonstrated that Hp, rather than impairing ApoE binding to β -amyloid, promotes the formation of the complex between β -amyloid and ApoE2, ApoE3, or ApoE4 (111). Hence, the suggestion from this study is that the risk of developing AD not only might be linked to the different ApoE isoforms, but also may rely on the level and type of Hp. Song et al. (112) reported a significantly higher level of serum Hp in patients with AD than in healthy control subjects. Their results support the hypothesis that oxidative stress is a key event in the pathogenesis of AD. The specific genotypes were not examined in this

study, so a future investigation will be needed to determine the impact of Hp1 versus Hp2 on AD pathogenesis. Brain atrophy, reduced cerebral glucose metabolism, and central nervous system insulin resistance are all features of AD, PD, and type 2 diabetes (113,114).

Finally, several studies have examined the effect of Hp genotype on the outcome and resolution of intracerebral hemorrhage. Murthy et al. (115) recently reported that patients with the Hp2-2 genotype experience worse outcomes after intracerebral hemorrhage. These results were supported by another study by Kantor et al. (116) in the context of subarachnoid hemorrhage, in which patient outcomes were worse in those with the Hp2-2 genotype.

Cardiology

Hp genotype has been consistently associated as a marker and risk factor for CVD. In a study by Chapelle et al. (117), patients with the Hp2-2 genotype had more severe myocardial infarction. Hp was later examined in a prospective study of >342,000 patients over 11.8 years. Hp was shown to be a significant risk factor of acute myocardial infarction (AMI), stroke, and heart failure (HF) (118). Data from this study established an association of Hp in these cardiovascular events that was stronger for men than for women. Hp was almost as predictive as total cholesterol for AMI and about as predictive as total cholesterol of stroke, with a stronger relationship to ischemic disease than to hemorrhagic stroke where a 4.2-fold increase in risk was observed in ischemic disease. Haas et al. (119) likewise established an association with Hp as a potential prognostic biomarker in AMI. In summary, Hp genotype carried as much predictive value as total cholesterol for risk of AMI and stroke.

HF and stroke also have been associated with Hp genotype (118,120). Costacou et al. (120) described a borderline increased risk

of stroke in people with type 1 diabetes and the Hp1-1 genotype. In an earlier study (121), this same group described an increased risk for CVD in people with the Hp2-2 genotype compared to those with the Hp1-1 genotype. Staals et al. (122) likewise suggested an increased risk for lacunar stroke depending on patients' Hp genotype. The aforementioned study by Holme et al. (118) established a twofold increased risk for stroke and a 1.5-fold increased risk for HF. Hp2-2 has been associated with higher total serum and free cholesterol concentrations, reduced graft survival in patients undergoing coronary artery bypass, and a greater risk of restenosis after stent implantation (Hp2-2 36% vs. Hp2-1 31%) (123–125).

Hp2-2 is a clear genetic risk factor for the development of aneurysm, coronary atherosclerosis, and unstable carotid stenosis independent of, or in connection with, many of the classical risk factors, including dyslipidemia, hypertension, diabetes, smoking, and hyperhomocystenemia (12,126,127). In one study involving male patients with diabetes, Lioupis et al. (128) demonstrated that patients had a higher serum level of homocysteine and that those with the Hp2-2 genotype had a higher concentration of iron in the atherosclerotic plaque. These findings suggest that increased intraplaque iron deposition may be responsible for increased oxidative stress and instability of the carotid plaque.

Additionally, Hp is a risk factor for developing refractory hypertension in patients with existing hypertension (129,130). Data demonstrate that patients with the Hp2-2 genotype require more antihypertensive therapy to control blood pressure than do patients with other genotypes. Patients with the Hp2-2 genotype also require more support and follow-up than do patients with alternate Hp genotypes (129,130). Conversely, patients with the Hp1-1 genotype have a lower rate of complications than do hypertensive patients

with either the Hp2-1 or the Hp2-2 genotype (130).

The literature strongly supports the hypothesis that the Hp2-2 genotype provides much less protection from oxidative damage to arteries in patients with atherosclerotic plaques. The Hp2-2 genotype also confers additional CVD risk, as well as other complications, including aneurysm, carotid plaque rupture, myocardial infarction, and decreased survival after coronary artery bypass. It has become clear in the past 20 years that Hp can be considered a predictor of susceptibility to cardiovascular disorders and a measure of patients' prognosis and outcomes.

Diabetes

The generation of ROS has a significant role in the generation of vascular complications in people with diabetes (61). Complications in people with diabetes resulting from HP genotype include cardiovascular events, retinopathy, and nephropathy. In studies of patients on hemodialysis, Levy and others (131,132) have described a protective effect in patients with the Hp1-1 genotype. Two independent studies by Levy et al. (133,134) showed that Hp genotype correlates with vascular complications. For example, the Hp1-1 genotype protects against vascular injury as a consequence of increased antioxidant activity compared to Hp2-1 or Hp2-2. Similar results were described by Szafranek et al. (135), who found that Hp was a major susceptibility gene for diabetic vascular complications. The Hp2 genotype has been reported to be associated with an increased risk for cardiovascular events such as myocardial infarction in individuals with type 2 diabetes (134,136). These studies and others show that Hp2 patients can have as much as a five times greater coronary disease risk and susceptibility to CVD compared to patients with the Hp1-1 genotype.

In addition to cardiovascular risk and complications, Hp genotype also correlates to increased risk for

diabetic retinopathy in people with type 2 diabetes. In patients with normal blood pressure, Mogarekar and Hampe (137) showed an increased risk for the development of severe retinopathy in those with the Hp2-2 genotype. When compared to other genotypes, this study showed a graded risk relationship with the number of Hp2 alleles.

Hp genotype determination, therefore, can be valuable in the assessment and management of diabetic retinopathy. Research has clearly demonstrated that blindness from diabetes is preventable with early diagnosis, clarification of risk factors, and timely photocoagulation where appropriate. Hp genotyping can allow for earlier identification of and prompt early intervention for patients who may suffer from diabetic retinopathy.

Diagnostic Implications of Hp

Plasma Hp concentrations have been examined for a variety of pathogenic disorders. Both high and low levels of Hp are indicative of clinical conditions. Elevated plasma concentrations have been described in cases of trauma, burn, inflammation, and cancer because of the acute-phase nature of the protein and its role in the removal of oxidative species from the circulation (33,61,138). Plasma levels are high beginning several days after the inflammatory or traumatic insult and return to normal within several weeks (139). Conditions in which Hp is decreased include malnutrition, hemolysis, hepatic disease, allergic reactions, and seizure disorders (139–142).

Given the involvement of Hp in the pathogenesis of cardiovascular, neurological, infectious, and inflammatory conditions, it would be beneficial to patients to have a diagnostic tool that can quickly identify Hp genotype so proper therapy can be implemented. For example, Hp2-2 is found in ~36% of the population. For people with this genotype, at least 10 studies examining nearly 175,000 patients collectively have sought to

determine the effects of this genotype versus nonhomozygous carriers.

A rapid test can be employed to initiate a therapy such as vitamin E or vitamin C to minimize the progression of CVD by as much as 30–40% (143). Initiation of 400 IU of vitamin E has been shown effective in reducing cardiovascular events (12,144). The antioxidant activity of vitamin E neutralizes the oxidative capacity of free heme and serves as a surrogate for Hp activity (145). Free heme can result in the oxidation of LDL particles and lead to the growth and instability of atherosclerotic plaque. Moderate doses of vitamin C and E can be effective in stabilizing oxidative stress and reducing the levels of circulating free radicals (12,145). The current methodology for determining genotype is gel electrophoresis, which is time-consuming and not optimal for large patient volume or commercial diagnostic purposes. A method developed by Levy et al. (146) uses an enzyme-linked immunosorbent assay (ELISA) to characterize the Hp genotype. This ELISA method is a user-friendly, accurate diagnostic tool for determining Hp genotype in the research environment but has not been employed in a commercial diagnostic capacity.

New methods for high-volume patient genotyping allow for rapid screening and early implementation of alternate antioxidant therapy and provide a tremendous benefit to people with diabetes.

Conclusion

The most thoroughly characterized property of Hp is its capacity to bind free Hb and promote endocytosis via the CD163 scavenger receptor and intracellular degradation of the Hp-Hb complex (9,37,147). In this process, Hp can reduce the loss of free Hb through glomerular filtration and promote the recycling of iron. In addition to its role in clearance, Hp has a protective function in that heme and iron released from free Hb

are removed from circulation, thereby preventing the production of ROS.

In the context of diabetes, the role of Hp can be either protective or pathogenic. Patients with the Hp2 allele have a significantly higher risk for cardiovascular, neurological, and infectious complications. Of particular concern in people with diabetes is the significantly increased risk for myocardial infarction, stroke, infection, and kidney disease in those with an Hp2-2 genotype.

Although the risk is higher for those with Hp2-2, there are several ways to reduce this risk back to baseline cardiovascular risk for patients with diabetes. The administration of exogenous antioxidants such as vitamin C and vitamin E can mitigate the deleterious effects of an Hp2-2 genotype (12,134,143,148,149). Thus, it is crucial for the research and diagnostic community to develop new methods of identifying patient genotype and advancing therapeutics for the treatment of people with diabetes who have the Hp2 polymorphism.

Duality of Interest

The authors are employed by MyGenetx, a company that performs diabetes laboratory testing. No other potential conflicts of interest relevant to this article were reported.

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