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Chapter 1

EXERCISE AND THE HDL QUALITY

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ABSTRACT

Circulating high-density lipoprotein (HDL) cholesterol (HDL-C) is a marker associated with cardiovascular health. Exercise is generally known to increase the HDL-C levels, and this can, in part, explain its cardioprotective effects. While HDL-C is a marker of the HDL quantity, special attention to the HDL quality (reflective of HDL functionality) has recently arisen as a new view on HDL biology and clinical studies of HDL. However, there is less information on the changes in the HDL quality induced by exercise. When we present some data regarding the association between exercise and the HDL quality, i.e., using oxidatively modified HDL and cholesterol efflux functionality of HDL, exercise may favorably affect the HDL quality. The present paper further encourages taking into consideration the view of HDL quality in relation to exercise, in addition to HDL-C.

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ASSOCIATION OF HIGH-DENSITY LIPOPROTEIN WITH CARDIOVASCULAR HEALTH

Lipid and lipoprotein disorders are relevant pathologic entities associated with cardiovascular health [1, 2]. Circulating high-density lipoprotein (HDL) cholesterol (HDL-C) is a well-known marker of cardioprotection; that is, relatively high HDL-C levels are inversely related to cardiovascular disease [3, 4]. Although a deeper understanding of HDL is still required, cardioprotection, as mediated by HDL, is explained by the following mechanisms: HDL exerts the efflux function of cholesterol from lipid-laden macrophages in the arterial wall and then delivers cholesterol to the liver, termed the reverse cholesterol transport pathway [5]. Cellular transporter molecules, such as adenosine triphosphate-binding cassette transporter A1, are involved in the step of cholesterol efflux achieved by HDL [5]. HDL inhibits the oxidation of atherogenic lipoproteins, such as low-density lipoprotein (LDL) [6] and exerts anti-inflammatory effects [7]. Various HDL-mediated mechanisms, including the inhibition of glycation, homocysteinylation, apoptosis and thrombosis, as well as the enhancement of nitric oxide, have also been documented [8-10]. These functions are, in part, due to the fact that HDL includes many protein molecules [7, 11].

ASSOCIATION OF EXERCISE WITH HDL-C

A habit of exercise, one of physical activities, is considered to favorably affect the development of lipid and lipoprotein disorders, and the effects of exercise on lipid and lipoprotein metabolism are best investigated by focusing on the HDL-C levels in particular [12, 13]. Well-summarized reviews have described that the HDL-C level increases 4-18% by exercise, while the LDL-cholesterol level is not changed by exercise [12]. Several lines of evidence follow these observations, such as the association of exercise with an increased HDL-C level, regardless of an older age, gender and various disease conditions [14-16]. Even though conflicting data exist, this phenomenon is thought to be due to the different characteristics and methodologies of exercise used across studies (i.e., intensity, amount, frequency, duration [acute/chronic/intermittent] and type [aerobic/resistance]) [12]. Thus, the general consensus is that exercise mildly-to-moderately but favorably moderates HDL metabolism, helping to promote cardiovascular health.

Exercise is currently recognized to be a cornerstone modality for improving cardiovascular outcomes.

HDL comprises a heterogeneous family of particles, differing in density, size, membrane charge and lipid and protein composition [11, 17]. Based on the subfraction (subclass/subpopulation) of HDL particles, the HDL-C level is basically divided into the HDL2-C (large-sized) and HDL3-C (small-sized) levels [17]. Evidence indicates that exercise increases the HDL2-C level [18]. Recent studies also highlight the finding of increased HDL2-C levels induced by exercise [19-21], possibly contributing to increased total HDL-C levels as a result of exercise.

PARADIGM SHIFT ON THE VIEW OF HDL

While the effects of the HDL subfraction on cardiovascular outcomes remain to be established [22], new insights have been obtained regarding the different roles of the subfraction of HDL in terms of cardioprotective properties [11, 23]. Surely, low HDL-C levels predict the development of cardiovascular disease, although interventional studies have revealed that the HDL level, as elevated by drugs such as cholesteryl ester transfer protein inhibitors, does not always protect against poor cardiovascular outcomes [24, 25]. Therefore, the associations between HDL and cardiovascular risks are recognized to be more complex than previously thought [7, 17, 23]. The total HDL-C levels express the nearly overall number of HDL particles; however, we think that the ‘quantity of HDL’ is not necessarily a perfect marker with respect to the evaluation of HDL in cardiovascular practice. Therefore, the principle of ‘HDL quality’ has arisen as a new concept ‘beyond HDL-C,’ which is considered to rely on the functionality of HDL, rather than HDL-C [23, 25-27]. Nonetheless, which measurements and/or tests are suitable for evaluating HDL and application in cardiovascular practice remains to be determined [28].

ASSOCIATION OF EXERCISE WITH OXIDIZED HDL AS A MARKER OF HDL QUALITY

Apolipoprotein A-I (apoA-I) is known to be modified at several specific residues (i.e., methionine and tyrosine residues) [29-31]. ApoA-I is thought to

be anti-oxidative, but more susceptible to oxidation than proteins on LDL [32-35]. The modification of apoA-I may be associated with 'HDL quality,' including the cholesterol efflux function of HDL [36]. As apoA-I is the most major protein component of HDL, the measurement of oxidatively modified apoA-I may be a marker of oxidized HDL (oxHDL).

We recently developed an ELISA system to measure the oxHDL levels [37, 38]. This assay utilizes an antibody against oxidized human apoA-I generated by treatment with H_2O_2, and does not react with native HDL, but rather a broad range of oxidative substances of HDL [37]. Our studies using this assay have shown increased oxHDL levels in patients with prediabetes and diabetes, an oxidative condition [37, 38]. Since no studies have characterized the relationship between exercise and oxHDL, we conducted a pilot study (with a single arm and small sample design) to see the changes in the oxHDL levels among 11 males (mean age 66 years) during a 6-week exercise intervention (Kotani K., unpublished data). In this population, while the overall HDL-C levels were reduced (this finding appears to be somewhat unexpected considering the above general consensus on the changes in HDL-C induced by exercise), the oxHDL levels tended to be reduced by increased exercise (Table 1). Of note, during the exercise intervention period, the changes in the HDL-C levels correlated inversely ($r = -0.62$, $p < 0.05$) with those in the levels of oxHDL/HDL-C, an index of HDL quality. Therefore, oxidatively modified HDL might have been improved in individuals with increased/sustained HDL-C levels during the study period.

Table 1. Changes of parameters at the pre- and post-intervention

Parameters	Pre	Post	P
Body mass index, kg/m^2	22.7 ± 1.6	22.9 ± 1.7	NS
Glucose, mg/dL	105 ± 11	100 ± 9	0.05
Total cholesterol, mg/dL	231 ± 40	226 ± 36	NS
Triglyceride, mg/dL	96 (66-111)	96 (76-134)	NS
HDL-C, mg/dL	66 ± 12	56 ± 8	< 0.01
OxHDL, U/mL	219 ± 56	178 ± 47	0.09
OxHDL/HDL-C	3.4 ± 0.9	3.2 ± 1.0	NS

Data are expressed as mean ± standard deviation or median (interquartile range).

NS: no significance, HDL-C: high-density lipoprotein cholesterol, oxHDL: oxidized high-density lipoprotein.

P: paired t-test (pre and post). Triglyceride values were log-transformed.

Recently, an additional marker of oxHDL was developed by other investigators, and that study showed high levels of oxHDL in patients with cardiovascular disease [40]. Research on the association of oxHDL with cardiovascular outcomes in relation to exercise is ongoing.

ASSOCIATION OF EXERCISE WITH CHOLESTEROL EFFLUX OF HDL AS A MARKER OF HDL QUALITY

Cholesterol efflux assays have been applied, and studies using these assays have shown the superior predictive value of cholesterol efflux functionality of HDL in predicting cardiovascular disease compared to the simple HDL-C levels [41, 42]. One study investigated the relationship between a 9-week exercise program (plus diet) and cholesterol efflux in obese females [43], showing non-significant changes in the cholesterol efflux functional levels, with a significant inverse correlation between weight loss and an increased cholesterol efflux functional level during the intervention period [43]. We conducted a pilot study (with a single arm and small sample design) to see the changes in the cholesterol efflux functional levels among 32 non-obese subjects (mean age 68 years) during a 6-week exercise intervention (Kotani K., Remaley A.T., unpublished data). This population was non-obese. A cholesterol efflux assay was performed based on a previous method [44]: that is, near confluent cells were labelled with ^{3}H-cholesterol for 48 hours, washed and effluxed for 18 hours with the indicated lipid acceptors prepared in α-MEM containing 1 mg/mL of BSA (α-MEM/BSA). The percentage efflux was calculated by subtracting the radioactive count in the blank medium from the radioactive count in the presence of the acceptor and then dividing the result by the sum of the radioactive count in the medium plus the cell fraction. As a result, the overall HDL-C levels increased, and the cholesterol efflux functional levels increased as exercise increased (Table 2). Considering the recent concept of the superiority of the cholesterol efflux functional levels in predicting cardiovascular disease to the simple HDL-C levels, it would be meaningful to note the exercise can improve and/or enhance cholesterol efflux functionality.

Table 2. Changes of parameters at the pre- and post-intervention

Parameters	Pre	Post	P
Body mass index, kg/m^2	22.9 ± 2.9	22.6 ± 3.1	< 0.01
Glucose, mg/dL	99 ± 15	98 ± 21	NS
Total cholesterol, mg/dL	215 ± 37	230 ± 41	< 0.01
Triglyceride, mg/dL	80 (55-112)	102 (64-124)	NS
HDL-C, mg/dL	64 ± 13	68 ± 15	< 0.01
Cholesterol efflux, %	21 ± 3	26 ± 3	< 0.01

Data are expressed as mean ± standard deviation or median (interquartile range).
NS: no significance, HDL-C: high-density lipoprotein cholesterol.
P: paired t-test (pre and post). Triglyceride values were log-transformed.

PERSPECTIVES AND CONCLUSION

Circulating HDL-C, represented as the HDL quantity, is a marker of cardioprotection, and exercise generally increases the HDL-C levels. The HDL quality (reflective of HDL functionality) has recently received much attention with respect to cardiovascular health. While less information is available on the association of exercise with HDL quality, we herein presented pilot data regarding the association between exercise and HDL quality based on oxidatively modified HDL and cholesterol efflux functionality of HDL, showing that exercise can favorably affect the HDL quality. This encourages practitioners to take into consideration the view of HDL quality in relation to exercise, besides HDL-C, in terms of the future direction of cardiovascular health. Further factors, such as genetic components, may potentially be considered in order to clarify the association between exercise and the biology of HDL quality [12]. In the near future, with assessments of the HDL quality, the ‘quality of exercise’ may also be discussed.

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