Effects of Pravastatin on the Expression of ATP-Binding Cassette Transporter A1

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ABSTRACT

In vitro inhibition of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase causes the suppression of liver X receptor (LXR) activity. Because LXR regulates the expression of ATP-binding cassette transporter (ABC) A1, which is involved in the high-density lipoprotein-related reverse cholesterol transport pathway, we examined the effects of an HMG-CoA reductase inhibitor pravastatin on ABCA1 expression in vitro and in vivo. Pravastatin (10 μM) significantly reduced the transcript levels of murine ABCA1 gene by 35% in RAW264.7 macrophages under a lipoprotein-deficient condition. The inhibition was due to the decreased mevalonic acid production because addition of exogenous mevalonic acid restored ABCA1 mRNA levels. In addition, cholesterol and 22(R)-hydroxycholesterol thoroughly blunted the inhibition. Furthermore, pravastatin did not decrease ABCA1 mRNA and protein levels in HepG2 hepatocytes even in the absence of exogenous LXR agonists. Oral dosing of pravastatin (0.1% concentration in drinking water) for 24 h or 2 weeks to mice did not decrease ABCA1 mRNA and protein levels in the liver and leukocytes. Interestingly, pravastatin significantly increased both hepatic and leukocyte LXRα mRNA levels. Thus, although HMG-CoA reductase inhibitors suppress ABCA1 mRNA expression in the absence of LXR agonists, in vivo inhibition of HMG-CoA reductase is unlikely to cause such suppression.

For over three decades, it is believed that high-density lipoprotein (HDL) protects against atherosclerosis through the action of the reverse cholesterol transport pathway (Glimset, 1968). In this pathway, HDL or its apolipoproteins mediate the removal of excess cholesterol from peripheral cells back to the liver for subsequent excretion into the bile. The recent discovery of the role of ATP-binding cassette transporter (ABC) A1 has given further support for the reverse cholesterol transport hypothesis. Loss of ABCA1 function leads to Tangier disease and familial HDL deficiency, which are genetic disorders characterized by the marked reductions in plasma HDL concentrations and increased risk of cardiovascular disease (Singaraja et al., 2003). Functional defect of ABCA1 in mice has also been reported to induce the absence of HDL (Christiansen-Weber et al., 2000; McNeish et al., 2000), and the inactivation of ABCA1 in macrophages has been shown to increase the susceptibility to atherosclerosis (Aiello et al., 2002; van Eck et al., 2002). Cells isolated from patients with Tangier disease are defective in the process of apolipoprotein-mediated removal of cholesterol and phospholipids (Walter et al., 1994; Francis et al., 1995; Rogler et al., 1995; Remaley et al., 1997). Therefore, it is assumed that ABCA1 transports intracellular cholesterol and phospholipids to cell surface-bound apolipoproteins and forms nascent HDL. Thus, ABCA1 seems to be essential for the first and rate-controlling step in the reverse cholesterol transport pathway.

Investigations on the regulation of ABCA1 gene expression have revealed that ABCA1 is induced in cholesterol-loaded cells as a result of the activation of the nuclear receptors liver X receptor (LXR)/retinoid X receptor heterodimer (Costet et al., 2000; Repa et al., 2000; Venkateswaran et al., 2000a). At least under such conditions, oxysterols seem to serve as the ligands for LXR (Forman et al., 1997; Venkateswaran et al., 2000a). Synthetic LXR agonists can elevate plasma HDL concentrations in experimental animals (Schultz et al., 2000; Cao et al., 2002; Greffhorst et al., 2002). Deficiency of LXR leads to exacerbation of atherosclerosis (Schuster et al., 2002; Tangirala et al., 2002), whereas a synthetic LXR agonist

ABBREVIATIONS: HDL, high-density lipoprotein; ABC, ATP-binding cassette transporter; LXR, liver X receptor; LDL, low-density lipoprotein; HMG-CoA, 3-hydroxy-3-methylglutaryl coenzyme A; DMEM, Dulbecco’s modified Eagle’s medium; LPDS, lipoprotein-deficient bovine calf serum; OHC, hydroxycholesterol; PCR, polymerase chain reaction; GAPDH, glyceraldehyde-3-phosphate dehydrogenase.
exerts antiatherogenic properties probably due to an induction of ABCA1 expression (Joseph et al., 2002). Thus, LXR is potentially a central mediator of ABCA1 action. LXR has two subtypes, LXRα and LXRβ, which are considered to regulate ABCA1 expression (Lund et al., 2003).

It has become common knowledge that the management of low-density lipoprotein (LDL) cholesterol concentrations is important for the prevention of coronary heart disease, and if necessary, treatment with 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitors for lowering LDL is recommended (National Cholesterol Education Program, 2001). HMG-CoA reductase inhibitors reduce LDL cholesterol concentrations through the blockade of mevalonate pathway and consequent increment of LDL receptor expression in the liver (Goldstein and Brown, 1990). Interestingly, Forman et al. (1997) reported that HMG-CoA reductase inhibition in the liver (Goldstein and Brown, 1990), suggesting that it was enough to reduce the metabolism of LDL cholesterol through the blockade of mevalonate pathway and consequent increment of LDL receptor expression in the liver (Goldstein and Brown, 1990). Interestingly, Forman et al. (1997) reported that HMG-CoA reductase inhibitors could inhibit constitutive activity of the LXRα-retinoid X receptor heterodimer by the reduction of mevalonic acid in vitro. Although ABCA1 is a notable target of these transcription factors, it remains to be determined whether HMG-CoA reductase inhibitors reduce expression levels of ABCA1. If they suppress ABCA1 activity, combination therapy with such an agent as LXR agonist, which increases ABCA1 expression (Lund et al., 2003), might be more effective for the prevention of atherosclerosis. To address this issue, we examined the effects of an HMG-CoA reductase inhibitor pravastatin on ABCA1 expression in vitro and in vivo.

Materials and Methods

Chemicals. Pravastatin sodium was kindly provided by Sankyo Co. (Tokyo, Japan). Cholesterol was obtained from Wako Pure Chemicals (Osaka, Japan). All other chemicals were purchased from Sigma-Aldrich (St. Louis, MO) unless otherwise stated.

Cell Culture. The murine macrophage cell line RAW264.7 and human hepatocyte cell line HepG2 were originally obtained from American Type Culture Collection (Manassas, VA). These cells were cultured in Dulbecco’s modified Eagle’s medium (DMEM) or DMEM/Ham’s F-12 mixture (In VitroGraft, Carlsbad, CA), supplemented with 10% heat-inactivated fetal bovine serum (Dainippon Pharmaceutical, Osaka, Japan), 100 U/ml penicillin, and 100 μg/ml streptomycin (In VitroGraft) at 37°C in an atmosphere of 5% CO₂, 95% air. For the present experiments, cells were seeded in six-well plates and grown to confluence within 2 to 4 days. When the cells became nearly confluent, the medium was replaced by DMEM with 10% lipoprotein-deficient bovine calf serum (LPDS; Biomedical Technologies, Stoughton, MA) and mevalonic acid (0.1 mM). After 24 h, RAW264.7 and HepG2 cells were then incubated for 8 h in DMEM supplemented with 10% LPDS + pravastatin (1 or 10 μM) in the presence of either vehicle (dimethyl sulfoxide), mevalonic acid (25 μM), or 22(R)-hydroxycholesterol (OHC) (25 μM), and total RNA was purified.

Mice and Treatments. Male C57BL/6 mice were obtained from Japan SLC (Hamamatsu, Japan) at 7 weeks of age and maintained under a specific pathogen-free condition with controlled temperature and humidity and a 12-h light/12-h dark cycle. Mice were given a standard laboratory diet (CE-2; CLEA Japan, Tokyo, Japan) and water ad libitum. After a 7-day acclimation period, animals were divided into three groups and given drinking water with or without pravastatin (0.01 or 0.1%) for 2 weeks. Blood and liver samples were obtained from mice after an 8-h fast (from 6:00 AM to 2:00 PM). All animal procedures were performed in accordance with the Guidelines for Animal Research at Jichi Medical School.

Serum Cholesterol Measurement. Serum total cholesterol was determined enzymatically using kits purchased from Wako Pure Chemicals.

RNA Extraction and Real-Time Quantitative PCR. The isolation of total RNA was achieved using the RNeasy Mini Kit or the QIAamp RNA Blood Mini kit according to the manufacturer’s instructions (QIAGEN, Valencia, CA). Reverse transcription was done by 1.2 μg of total RNA, random hexamer primer, and RevertAid M-MuLV reverse transcriptase (Fermentas, Hanover, MD). The resulting cDNA equivalent to 60 ng of RNA was used for the real-time quantitative PCR in the ABI Prism 7700 sequence detection system (Applied Biosystems, Foster City, CA). All of specific sets of primers and TaqMan probes in the present study were obtained from Applied Biosystems (Assays-on-Demand Gene Expression Products and TaqMan Rodent GAPDH Control Reagents). All primer sets but that of TaqMan Rodent GAPDH Control Reagents were designed to be located in two exons to avoid the amplification of potentially contaminating genomic DNA. To control for the variation in the amount of DNA available for PCR in the different samples, gene expressions of the target sequence were normalized in relation to the expression of an endogenous control, glyceraldehyde-3-phosphate dehydrogenase (GAPDH). In the present study, no agent affected the mRNA expression of GAPDH: the coefficients of variation of its Ct values were all <3%. Because the efficiency of the target amplification was approximately equal to that of the GAPDH amplification, data were analyzed using comparative threshold cycle method (Su et al., 2002). Because the intra- and interassay coefficients of variation of the relative expression values were ~20%, we considered the mean relative values of less than 0.8 or more than 1.2 to be significant in this study.

Western Blot Analysis. Cells and tissue samples were lysed in Celllytic MT reagent supplemented with 1% protease inhibitor cocktail. The supernatant was collected after centrifugation and the protein concentrations were determined using the DC Protein Assay kit (Bio-Rad, Hercules, CA). Proteins (40 μg/lane) were separated by SDS-polyacrylamide gel electrophoresis (6% Tris-glycine gel; Invitrogen) and then transferred to polyvinylidene difluoride membranes (Invitrogen). Nonspecific binding sites were blocked with 5% skim milk in Tris-buffered saline containing 0.1% Tween 20. Membranes were subsequently incubated for 1 h at room temperature with anti-human ABCA1 rabbit polyclonal antibody (Novus Biologicals, Littleton, CO). The antibody, which reacts with both human and mouse ABCA1, was diluted 1000-fold in Tris-buffered saline containing 0.1% Tween 20 and 3% skim milk. After incubation with a secondary horseradish peroxidase-labeled antibody, the blots were visualized using ECL Plus Western Blotting Detection Reagents (Amersham Biosciences Inc., Piscataway, NJ). Quantification of the signals was performed by densitometry using the 1D Image Analysis Software (Eastman Kodak, Rochester, NY).

Statistics. Data were analyzed using the Mann-Whitney U test or one-way analysis of variance with a post-test of Fisher’s protected least significant differences. Values are presented as the means ± S.E., and a P value of less than 0.05 was considered significant. All calculations were performed using the computer program StatView, version 5.0 (SAS Institute, Cary, NC).

Results

Effects of Pravastatin on Abca1 mRNA Expression Levels in RAW264.7 Cells. To investigate whether an HMG-CoA reductase inhibitor reduces mRNA expression levels of ABCA1 in vitro, RAW264.7 murine macrophages were cultured for 8 h in DMEM supplemented with 10% LPDS with or without pravastatin. Ten but not 1 μM pravastatin significantly increased transcript levels of HMG-CoA synthase 1, one of sterol regulatory genes (Goldstein and Brown, 1990), suggesting that it was enough to reduce the metabo-
lites of mevalonate pathway (Fig. 1A). As shown in Fig. 1B, 10 μM pravastatin reduced the transcript levels of murine ABCA1 gene Abca1 by 35%. The inhibition was due to the decreased mevalonic acid production because the addition of exogenous mevalonic acid could restore Abca1 mRNA levels (Fig. 1B). In addition, mevalonic acid itself significantly increased its mRNA levels. Cholesterol also significantly increased Abca1 mRNA levels and compensated for the inhibition by pravastatin. Moreover, the transcript levels of Abca1 were induced >100-fold in response to the LXR agonist 22(R)-OHC (Fig. 1B). Figure 1C shows the changes in mRNA expression levels of Abcg1, the gene of another member of ABC transporter superfamily, which is also known to be regulated by LXR (Venkateswaran et al., 2000a,b). In concordance with the results of Abca1, 10 μM pravastatin suppressed Abcg1 mRNA expression by 26% and did not diminish its 6-fold increase by 22(R)-OHC. On the other hand, pravastatin did not affect mRNA levels of Abca5, another member of ABCA subclass (data not shown). Because macrophages strongly express LXRα subtype (Lund et al., 2003), we also examined the changes in LXRα expression levels. Whereas pravastatin did not obviously alter LXRα expression itself, 22(R)-OHC induced a 1.3-fold increase (Fig. 1D). These results suggest that inhibition of HMG-CoA reductase decreases ABCA1 mRNA expression levels without affecting LXRα transcript levels in the absence, but not in the presence of LXR agonists.

**Effects of Pravastatin on ABCA1 mRNA Expression Levels in HepG2 Cells.** Whereas macrophage ABCA1 may be important for the prevention of atherosclerosis (Aiello et al., 2002; van Eck et al., 2002), hepatic ABCA1 may contribute to overall plasma HDL concentrations (Haghpassand et al., 2001; Basso et al., 2003). Therefore, we next investigated the effect of pravastatin in human hepatocytes, HepG2. As shown in Fig. 2A, treatment with pravastatin resulted in a concentration-dependent increase in HMG-CoA reductase mRNA levels. However, contrary to the results in RAW264.7 cells, the treatment did not induce any change of ABCA1 mRNA levels (Fig. 2B). In addition, pravastatin did not affect LXRα mRNA expression levels in HepG2 cells (Fig. 2C).

**In Vivo Effects of Pravastatin on Abca1 mRNA Expression Levels in Mice.** To confirm the above-mentioned findings in vivo, pravastatin was given to mice at 0.01 or 0.1% concentration in drinking water for 2 weeks. Total pravastatin dosages estimated by water consumption were about 20 and 200 mg/kg body weight/day in 0.01 and 0.1% pravastatin groups, respectively. In mice, the lower dosage of pravastatin is high enough to have various pleiotropic effects, although it has no effects on plasma lipid profile (Narisawa et al., 1994; Kwak et al., 2003; Sasaki et al., 2003). Under our condition, neither pravastatin treatment significantly reduced serum total cholesterol concentrations (control, 86.2 ±
6.3 mg/dl; 0.01% pravastatin, 88.5 ± 6.2 mg/dl; 0.1% pravastatin, 81.6 ± 2.6 mg/dl; n = 4–5 in each group). However, 0.1% but not 0.01% pravastatin significantly increased hepatic mRNA levels of HMG-CoA synthase 1 (Fig. 3A). Consistent with the in vitro findings of HepG2 cells, pravastatin treatment did not affect Abca1 transcript levels in the livers (Fig. 3B). Moreover, 0.1% pravastatin significantly increased LXRα mRNA levels (Fig. 3C). In the leukocytes, 2-week treatment with 0.1% pravastatin did not significantly affect either transcript levels of HMG-CoA synthase 1 (control, 1 ± 0.07; pravastatin, 1.06 ± 0.07; n = 4 in each group) or Abca1 (control, 1 ± 0.06; pravastatin, 1.05 ± 0.12), whereas the drug increased LXRα mRNA levels (control, 1 ± 0.06; pravastatin, 1.38 ± 0.10; P < 0.05).

Because it remained possible that some of the effects of pravastatin observed in vitro were transient, we further investigated the effects of 24-h treatment with 0.1% pravastatin. Serum total cholesterol concentrations did not differ between the two groups (control, 73.5 ± 1.9 mg/dl; pravastatin, 72.3 ± 4.3 mg/dl; n = 4 in each group). As shown in Fig. 4A, the treatment still did not significantly increase hepatic HMG-CoA synthase 1 mRNA levels, but it had already increased LXRα transcript levels. Interestingly, pravastatin tended to increase hepatic Abca1 mRNA levels. In the leukocytes, 24-h pravastatin treatment also increased LXRα mRNA expression without affecting Abca1 mRNA levels (Fig. 4B).

Effects of Pravastatin on ABCA1 Protein Levels. We further investigated whether pravastatin affects protein levels of ABCA1. In RAW264.7 cells cultured without 22(R)-OHC, ABCA1 level was very low, and obvious effect of pravastatin was not detected (Fig. 5A). Moreover, in concordance with the effects on mRNA expression, pravastatin did not affect ABCA1 mass in RAW264.7 cells cultured with 22(R)-OHC (Fig. 5B) nor in HepG2 cells (Fig. 5B). Furthermore, oral dosing of pravastatin for 2 weeks to mice did not affect hepatic or leukocyte ABCA1 protein levels (Fig. 6).

**Fig. 3.** Effects of 2-week pravastatin treatment on hepatic mRNA expression levels of HMG-CoA synthase 1 (A), Abca1 (B), and LXRα (C) in mice. Male C57BL/6 mice were divided into three groups at 8 weeks of age. These animals were given drinking water with or without pravastatin (0.01 or 0.1%) for 2 weeks, and thereafter mRNA expression levels of the target genes in the whole-liver were determined by the real-time quantitative reverse transcription-PCR. Data are means ± S.E. of four to five mice in each group and expressed as relative value to control. *P < 0.05, #, P < 0.01% pravastatin.

**Fig. 4.** Effects of 24-h pravastatin treatment on mRNA expression levels of HMG-CoA synthase 1, Abca1, and LXRα in the liver (A) and the leukocytes (B). Male C57BL/6 mice were divided into two groups at 8 weeks of age. These animals were given drinking water with (closed column) or without (open column) 0.1% pravastatin for 24 h, and thereafter hepatic and leukocyte transcript levels of the target genes were determined by the real-time quantitative reverse transcription-PCR. Data are means ± S.E. of three to four mice in each group and expressed as relative value to control. *, P < 0.05, #, P < 0.1 versus control.

**Fig. 5.** Effects of pravastatin on ABCA1 protein levels in RAW264.7 (A) and HepG2 cells (B). Cells were cultured for 48 h in DMEM supplemented with 10% LPDS ± pravastatin (10 μM) in the presence or absence of 22(R)-OHC (25 μM). Western blot analysis of ABCA1 was performed as described under Materials and Methods. Equal quantities of protein (40 μg) were run in each lane. Samples from one representative experiment are shown. Data are means ± S.E. of three independent experiments and expressed as relative value to control. *, P < 0.05 versus control.
proliferators-activated receptor

tase inhibitors have been shown to activate peroxisome
duced by the decrease of mevalonic acid. HMG-CoA reduc-
tin might compensate the suppression of LXR activity in-
expressed as relative value to control.

Fig. 6. Effects of pravastatin treatment on ABCA1 protein levels of the
liver (A) and the leukocytes (B) in mice. Male C57BL/6 mice were divided
into two groups at 8 weeks of age. These animals were given drinking
water with or without 0.1% pravastatin for 2 weeks, and thereafter
hepatic and leukocyte ABCA1 protein levels were determined by the
Western blot analysis. Equal quantities of protein (40 μg) were run in
each lane. Data are means ± S.E. of three mice in each group and
expressed as relative value to control.

Discussion

HMG-CoA reductase inhibitors, which are widely used in
the treatment of hypercholesterolemia, not only reduce LDL
cholesterol but also increase HDL cholesterol (National Cho-
esterol Education Program, 2001). The increase in HDL is
reported to be accompanied by the enhanced production of
apolipoprotein A-I (Schaefer et al., 1999). Therefore, HMG-
CoA reductase inhibitors seem to activate the reverse choles-
terol transport pathway, especially at the first step. On the
other hand, the previous studies clearly demonstrated that in
vitro inhibition of HMG-CoA reductase causes the suppress-
ion of LXR activity (Forman et al., 1997). Because LXR
regulates the expression of ABCA1, which is essential for the
first step of the reverse cholesterol transport pathway, we
examined the effects of HMG-CoA reductase inhibitors on
ABCA1 expression.

Consistent with the findings of the effects on LXR activity,
this study showed that pravastatin suppresses ABCA1
mRNA expression in RAW264.7 macrophages in the absence
but not in the presence of an LXR agonist. Contrary to the
results in RAW264.7 cells, pravastatin did not decrease
ABCA1 mRNA levels in HepG2 hepatocytes. Moreover, in
vivo treatment with pravastatin did not suppress either he-
patic or leukocyte ABCA1 mRNA expression in mice. The
precise mechanism that HMG-CoA reductase inhibition does
not decrease ABCA1 mRNA levels in HepG2 cells or in vivo
is not clear, but the present and previous results suggest the
following mechanisms.

First, the increase in LXRx expression levels by pravasta-
tin might compensate the suppression of LXR activity in-
duced by the decrease of mevalonic acid. HMG-CoA reduc-
tase inhibitors have been shown to activate pereoxisome
proliferators-activated receptor α (Martin et al., 2001) and γ
(Fajas et al., 1999). Ligand activation of either nuclear recep-
tor can lead to primary induction of LXRx and subsequent
induction of ABCA1 (Chawla et al., 2001; Chinietti et al.,
2001). The results obtained in this study showed for the first
time that in vivo treatment with pravastatin induces LXRx
mRNA expression.

Second, the decrease of geranylgeranyl pyrophosphate, one
of the major products of mevalonate pathway, by HMG-CoA
reductase inhibition may be involved in the changes of
ABCA1 mRNA expression. It has been reported that gera-
ylgeranyl pyrophosphate can reduce ABCA1 expression by
acting as an LXR antagonist (Forman et al., 1997; Gan et al.,
2001) and through the activation of Rho proteins (Gan et al.,
2001). It is possible that the depletion of geranylgeranyl
pyrophosphate might obviously affect ABCA1 expression in
presence but not in absence of LXR agonists.

Finally, there may be enough endogenous LXR agonists in
HepG2 cells and in vivo, such as oxysterols, to compensate
the decrease of mevalonic acid. In fact, ABCA1 mass in
HepG2 cells or in vivo was more than that of RAW264.7 cells
(cf. Figs. 5 and 6). In RAW264.7 cells, not only 22(R)-OHC but
also cholesterol compensated the effects of HMG-CoA re-
ductase inhibition on Abca1 and Abcg1 transcript levels (Fig. 1,
B and C). Denis et al. (2003) also reported that cholesterol
could increase ABCA1 expression levels probably through
the formation of hydroxysterols in fibroblasts. Because both
macrophages in atherosclerotic lesions and hepatocytes are
relatively rich in cholesterol and oxysterols, it does not seem
likely that HMG-CoA reductase inhibitors suppress ABCA1
expression in those cells that are main components in the
reverse cholesterol transport pathway. Moreover, ABCA1
mRNA levels in RAW264.7 cells were less than 1% of those in
the murine leukocytes (data not shown), and its protein lev-
els were very low without induction by LXR agonists (Fig. 5A).
Accordingly, the effects of HMG-CoA reductase inhibitors
in lowering ABCA1 expression from very low to even lower
under these conditions might not be important.

Currently, LXR-activating therapy is expected to decrease
atherosclerosis also in human (Lund et al., 2003). Adminis-
tration of synthetic LXR agonists actually can inhibit the
development of atherosclerosis in mice, but it also causes
fatty liver and hypertriglyceridemia at least partly by the
induction of sterol regulatory element binding protein-1c
(Lund et al., 2003). Therefore, some strategies for dissociat-
ing antiatherosclerotic from triglyceride raising effects are
needed to use LXR agonists for the treatment or prevention
of cardiovascular disease. On the other hand, many previous
data demonstrate both efficacy and safety of HMG-CoA re-
ductase inhibitors and further suggest that they have not
only lipid-lowering but various pleiotropic effects such as on
macrophages (Ando et al., 2000, 2003; Ota et al., 2003).
Because LXR agonists may also have various effects, it must
be carefully evaluated whether addition of ABCA1-activating
therapy to HMG-CoA reductase inhibitor treatment im-
proves the outcome of patients with cardiovascular disease.

In conclusion, although the inhibition of HMG-CoA re-
ductase suppressed ABCA1 mRNA expression in RAW264.7
macrophages in the absence of LXR agonists, pravastatin did
not reduce its levels in the macrophages cultured with LXR
agonists or HepG2 hepatocytes. Oral dosing of pravastatin
to mice did not suppress either hepatic or leukocyte ABCA1
expression. It is unlikely that pravastatin has adverse effects
on the action of the reverse cholesterol transport pathway by
inhibiting ABCA1 expression.

References

Ayaliel B, Brees D, Bourassa PA, Boyer L, Lindsey S, Coskran T, Haghpassand M,
and Francone OL (2002) Increased atherosclerosis in hyperlipidemic mice with
637.

affect circulating levels of soluble TNF receptor 2 in hypercholesterolemic pa-
tients? Atherosclerosis 166:413–414.


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