Pdf
LIPID CASE 252 Particle Core Composition

Hi Lipidaholics: The following case demonstrates a topic I have wanted to discuss in depth. It illustrates the concept of particle composition meaning what is being trafficked within the various lipoproteins? I was contacted by a nonprofessional who knows me and asked for a comment on their father's lipid profile.

The father (late 60s), who has difficult to control hypertension (on metoprolol, clonidine, hydralazine and furosemide) takes pravastatin 80 mg and has the following profile. He is also on omeprazole 20 mg and omega-3 krill oil. He does not have known atherosclerosis.

TC = 152 TG = 170 HDL-C = 37 VLDL-C = 34 LDL-C = 81
Non-HDL-C = 115 TC/HDL-C = 4.1 TG/HDL-C = 4.6

I was asked for my thoughts.

DAYSPRING DISCUSSION:

I want to start off by encouraging all of you obtain and read a classic paper that is really must reading for all who desire to understand atherogenesis. Thanks to my pal and fellow lipidologist Jamie Underberg for reminding me of this paper of which I had forgotten about.

Subendothelial Lipoprotein Retention as the Initiating Process in Atherosclerosis: Update and Therapeutic Implications Circulation. 2007;116:1832-1844. by Ira Tabas, et al. They state: ***"The key initiating process in atherogenesis is the subendothelial retention of apolipoprotein B–containing lipoproteins. Local biological responses to these retained lipoproteins, including a chronic and maladaptive macrophage and T-cell– dominated inflammatory response, promote subsequent lesion development."***

In other words atherosclerosis is a lipoprotein mediated, specifically an apoB lipoprotein disease. The "criminals" or sterols are brought to the scene of the crime as passengers inside of lipoproteins, specifically the apoB-containing lipoproteins. Quoting Fredrickson, Levy and Lees in the 1967 New England Journal classic: *"All abnormalities in plasma lipid concentrations or dyslipidemia can be translated into dyslipoproteinemia --- the shift of emphasis to lipoproteins offers distinct advantages in recognition and management of such disorders."*

Finally in the Journal of Internal Medicine 2006; 259: 247–258 a statement entitled *Apo B versus cholesterol in estimating cardiovascular risk and in guiding therapy: report of the thirty-person/ten country panel* nailed it by stating: "*all of the national and transnational screening and therapeutic guidelines are based on total or LDL cholesterol. This presumes that cholesterol is the most important lipoprotein-related proatherogenic risk variable. On the contrary, risk appears to be more directly related to the number of circulating atherogenic particles that contact and enter the arterial wall*

than to the measured concentration of cholesterol in these lipoprotein fractions. Each of the atherogenic lipoprotein particles contains a single molecule of apolipoprotein (apo) B and therefore the concentration of apo B provides a direct measure of the number of circulating atherogenic lipoproteins. Evidence from fundamental, epidemiological and clinical trial studies indicates that apo B is superior to any of the cholesterol indices to recognize those at increased risk of vascular disease and to judge the adequacy of lipid-lowering therapy. ***On the basis of this evidence, we believe that apo B should be included in all guidelines as an indicator of cardiovascular risk."***

So of course I advised a lipoprotein analysis in this patient, namely the NMR LipoProfile (nuclear magnetic resonance spectroscopy). A measured apoB would also suffice. LipoScience uses the Multi-Ethnic Study of Atherosclerosis (MESA) for lipoprotein distributions in a heterogeneous population. One can compare a given concentration in individual patients to those in the overall population (using population cutpoints percentiles).

The lipoprotein results were:

Lipid Panel: (all in mg/dL)
TC = 207 LDL-C = 110 HDL-C = 54 TG = 217 VLDL-C = 43 Non-HDL-C = 153
TC/HDL-C = 3.8 TG/HDL-C = 4.0

Total LDL-P = 1924 nmol/L (the 95th population percentile cutpoint using the MESA study - this means 95% of people would have a lower concentration - thus this is a very high risk level)
Small LDL-P = 1018 nmol/L (normal would be < 600)

Metabolic Syndrome Markers: The following parameters are used as markers of insulin resistance (and nothing else)! Changing the following parameters are not goals of therapy which for whatever reason far too many think they are.

Large VLDL-P = 11.0 nmol/L (quite high)
LDL Particle Size is 20.7 nm (with NMR technology particles > 20.5 nm are considered normal sized or Pattern A)
Large HDL-P = 2.7 mol/L (a very low level) DO NOT CONFUSE large HDL-P with Total HDL-P (which would add additional insight).

Since this panel was done at Labcorp, it is a truncated report (compared to what one would get if the specimen was sent to LipoScience in Raleigh). Missing is the total HDL-P plus other markers of insulin resistance. I have reason to believe that in the near future Labcorp may be increasing the number of NMR values on their reports.

Let's begin with standard risk evaluation as suggested by NCEP ATP-III (which does not use lipoprotein but rather lipid concentrations): If the above lipid panels were in a drug naive patient, then at first glance it might be easy to miss risk as many just zero in on the LDL-C of 81 or even 110 mg/dL. However, the history of hypertension combined with the lipid values establish the presence of the metabolic syndrome which increases his overall risk. Since he has at least two major risk factors (male, age, hypertension) he

would qualify for Framingham risk scoring. Using the first lipid panel along with age and hypertension his ten year risk is 20% (a coronary heart disease risk equivalent). Using the second profile his 10 year risk is 16% which would because of the number (>2) of risk factors present would put him in the moderately high risk category. Thus his LDL-C and non-HDL-C goals respectively would be < 100 and < 130 for a high risk patient or less than 130 and 160 (with an option for 100 and 130) for a moderately high risk person. Since he is not a very high risk patient, there is no NCEP option to set an LDL-C and non HDL-C goals of 70 and 100 mg/dL. Thus, the patient is pretty much at NCEP lipid goals: he is at goal using lipid profile 1 and darn close in lipid panel 2.

The reality is as follows: Unfortunately the above profiles are on-treatment values and NCEP risk scoring was never intended (and this is stated within the actual guidelines) to ascertain risks in persons on drug treatment. All of the epidemiologic data used to compute risk was from studies of patients not on drugs. Without knowing what his drug naive lipid profile is we cannot accurately know whether prior to therapy he was at moderate, moderately high or high risk although we might all guess it is the latter. Using NCEP criteria there is certainly no way to call him very high risk. Therefore, all we can say using the above lipid profiles is that pravastatin has pretty much achieved NCEP's lipid goals of therapy namely an LDL-C < 100 mg/dL and non-HDL-C < 130 mg/dL for a high risk patient or optional goals for a moderately high risk patient.

If we simply jump to the lipoprotein concentrations the LDL-P is greater than the 90th percentile of MESA and is in the very high risk category P (and recall that because of its 2-3 day half-life, 90% of the apoB particles are LDLs). So despite the at goal or at best borderline LDL-C and non-HDL-C concentrations, the man needs more aggressive therapy. He needs a more potent statin and even with that it is most unlikely an LDL-P of < 1000 nmol/L (the goal). Realize he has a grossly abnormal LDL-P on pravastatin 80 - he needs Crestor 40 mg and likely needs another additional medication and the potential options would be because of the high TG a fibrate (or fibric acid), Zetia 10 mg (ezetimibe), high dose prescription N-3 fatty acid (Lovaza) or Niaspan (extended release niacin) titrated to 2000 mg over time or even 3 of the above..

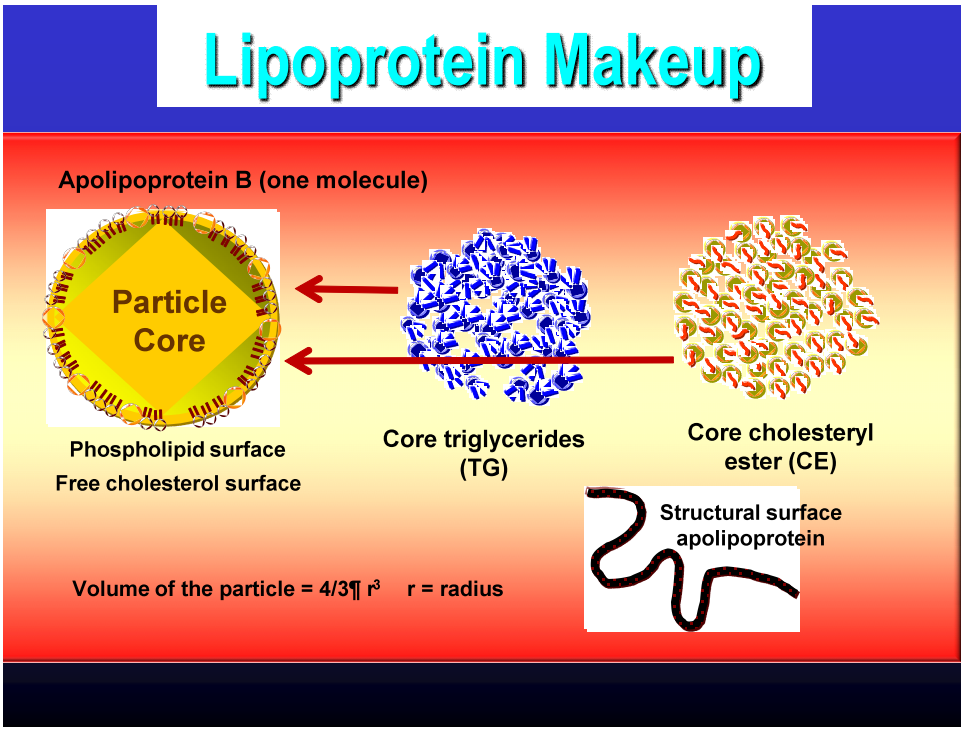
I want to use the rest of the newsletter explaining how the patient can have such an incredibly high LDL-P but also have normal or borderline LDL-C and non-HDL-C values. What seems especially perplexing is why in the face of high TG are his LDLs large? Would not you expect small LDLs in a metabolic syndrome patient with high TG?

The only way one can have a high LDL-P in the face of normal lipid surrogates of apoB or LDL-P (namely LDL-C and non-HDL-C) is to have cholesterol depleted particles. What does that mean? It means that for some reason the particle is trafficking less cholesterol than it should be. Let's examine the possibilities of how that is possible.

When looked at as a circular particle all lipoproteins have a volume which can be calculated as $4/3\ \pi\ \text{radius}^3$ (cubed). Because of the third power in the formula, larger particles have significantly higher volumes than do small. Thus if one only looked at size, compared to large LDL particles small LDLs likely traffic less cholesterol and in general the small LDL would be cholesterol depleted compared to the larger. Seemingly it would

take more small LDLs to traffic a given level of LDL-C than large LDLs. That is a general rule but there is a major exception and it must be present in this case.

When looked at in cross section, lipoprotein structure is a one molecule thick (monolayer) surface of unesterified cholesterol or UC (specifically called 3-hydroxy-cholesterol) mixed with phospholipids (PL). Both PL and UC are amphipathic molecules with the polar (dissolvable in water) end of the molecules on the exterior surface (interfaces with aqueous plasma) and non-polar (not dissolvable in water) end on the interior surface pointing towards the inner hydrophobic lipoprotein core. Thus apart from the surface layer, the other crucial part of the lipoprotein is its core because that is where the TG and cholesteryl ester or CE reside (a long chain fatty acid such as oleic acid replaces the -OH group at the # 3 position making CE or more accurately cholesterol-oleate much more hydrophobic or nonpolar than is unesterified cholesterol). Nothing else is inside the core of any lipoprotein except CE and TG. The makeup or composition of the particle core has everything to do with how many particles will be needed to traffic a given level of cholesterol.



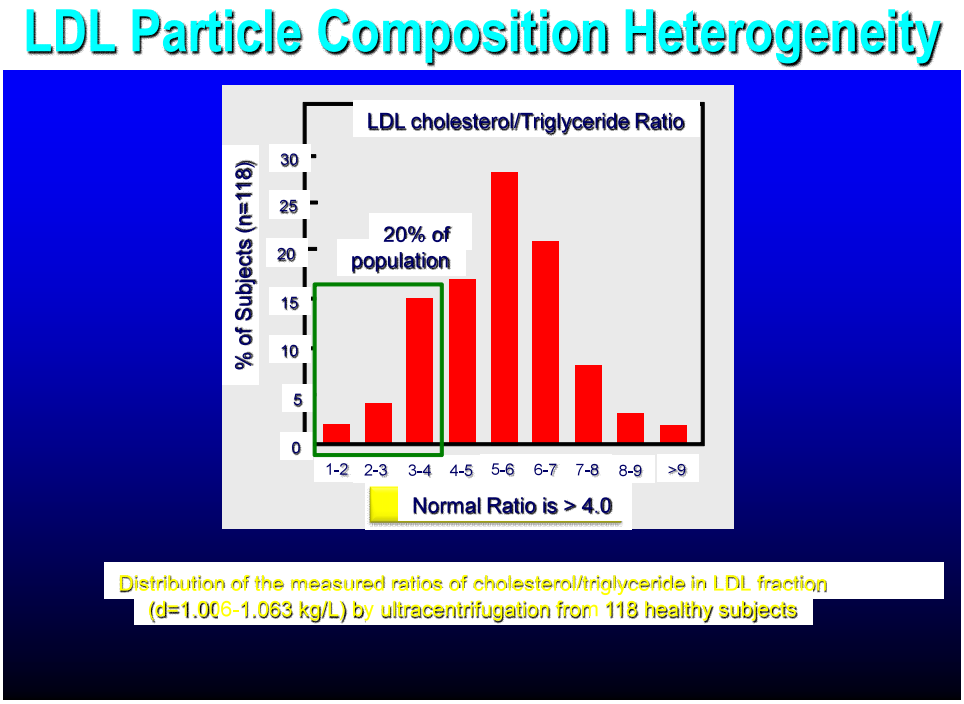
A normally composed LDL particle has a CE/TG ratio of 4 - i.e. a normal LDL particle carries 4 times more CE than it does TG (i.e. 80% CE and 20% TG). Any LDL particle that has a CE/TG ratio < 4.0 is a cholesterol-depleted (CE-poor) or a TG-rich LDL particle or both. The higher the ratio, the more cholesterol rich and TG-poor the particle is. We of course can measure LDL size in the laboratory but we cannot measure TG content of LDL (outside of research labs). Yet the patient under discussion has a very high LDL-P in the face of a normal sized (large or Pattern A) and an unremarkable LDL-C [the cholesterol carried within all of the LDL particles that exist in a deciliter (dL) of

plasma]. The obvious reason is the large LDL particle is carrying something else other than CE.

At a given LDL size the only other parameter that can influence the particles core CE content is TRIGLYCERIDES. Any LDL particle that is carrying more than the normal 20% TG in its core HAS TO BE CHOLESTEROL (CE) DEPLETED or if it is not CE-depleted it has to be large (as it would carrying excess TG and CE). Is it not unfortunate that labs cannot provide us with LDL-TG levels? In reality LDL-TG correlates better with elevated apoB or LDL-P than does LDL-C for the simple reason that anyone with an elevated LDL-TG level would likely have CE-depleted LDLs and it takes more CE-depleted particles to traffic a given level of LDL-C than CE-rich particles. So whether one has large or small LDLs, if they are carrying a < 4:1 CE/TG ratio (i.e. they are packing extra TG), they will be cholesterol depleted. So what if the patient under discussion with large LDL particles had an abnormally low CE/TG ratio (<4.0)? This would explain how a patient can have large numbers of large LDLs in the face of a normal LDL-C (the particles are pathologically trafficking TG instead of CE). In the case at hand the abnormal core composition, not the LDL size explains LDL-P / LDL-C disconnect or discordance. Unfortunately risk better follows LDL-P more than it does LDL-C. We have to stop relying on LDL-C as the sole determinant of risk and as the sole goal of therapy.

In the little known The Ludwigshafen Risk and Cardiovascular Health Study they showed that "alterations of LDL metabolism characterized by high LDL-TG are related to CAD, systemic low-grade inflammation (elevated hs-CRP), and vascular damage. High LDL-TGs are indicative of CE-depleted LDL, elevated IDL, and dense LDL. **LDL-TG may better reflect the atherogenic potential of LDL than LDL-C."** (Circulation. 2004;110:3068-3074.) Risk began with LDL-TG levels of 53 mg/dL. They also showed: "*First, LDL-TG predicts stable CAD independently of LDL-C. Second, in the MS and DM, LDL-TGs increase while LDL-C decreases. Third, systemic markers of low-grade inflammation are elevated at high LDL-TG but not at high LDL-C. Fourth, LDL-TG but not LDL-C is positively related to vascular adhesion molecules. Fifth, high LDL-TG is associated with high concentrations of VLDL, IDL, and dense LDL."*

In another study entitled Measurement Issues Related to Lipoprotein Heterogeneity, Otvos et al (Am J Cardiol 2002;90(suppl):22i–29i) have shown: *"The measured ratios of cholesterol/triglyceride in the LDL of 118 healthy subjects ranged from 1.8 to 11.5, and the distribution is shown in the graphic below. The majority (65%) of the study population had large LDL particles of "normal" composition (cholesterol/triglyceride ratio >4).14 However, a surprisingly large percentage of subjects (21%) had LDL particles that were compositionally cholesterol depleted (cholesterol/triglyceride ratio < 4) compared with normal. Even the most accurate LDL cholesterol measurement will underestimate by about 10% to 25% the actual amounts of LDL these individuals have in their bloodstream, compared with those with LDL particles containing a normal amount of cholesterol. Or put another way, measured LDL cholesterol values, even for people with LDL particles of the same size, can easily vary by 10 to 40 mg/dL without there being any difference in LDL particle concentration (or CAD risk, we would argue)."*



The next question is where do the LDLs acquire their cholesterol and TG: Since an LDL is a lipolytic by-product of an VLDL or IDL (i.e. a VLDL that has lost its TG) it is usually a TG-poor, CE rich particle. So the biggest source of the cholesterol within LDLs is the parent VLDL. However if the VLDLs are very TG-rich as they so often are in insulin resistant patients (note the elevated large VLDL-P in this patient) or if the large TG-rich VLDLs have a prolonged plasma residence time (delayed lipolysis meaning hydrolysis of or loss of TG) a lipid transfer protein is activated that swaps TG for CE molecules (a 1:1 swap) between CE-rich and TG-rich particles. That protein is called apolipoprotein D, better known as cholesteryl ester transfer protein (CETP). This man has increased numbers of large TG-rich VLDLs which utilizing CETP are sending their TG over to any CE-rich particles (LDLs and HDLs). The CE-rich particles accept the TG, but send back their CE to make room for the TG. LDLs that are accepting TG and losing CE become CE-depleted as well as TG-rich and will obviously have a low CE/TG ratio. Normally TG-rich, CE particles lose their TG in hepatic sinusoids where they are exposed to hepatic lipase (HL) and become small LDLs. One cause of patients with high TG having large LDL particles is HL deficiency. Normally HL removes TG from LDLs making them smaller in size. Patients with HL deficiency also have large HDLs with high HDL-C (obviously not the case with the patient under discussion).

So the lesson learned (hopefully): Particle core composition can play a major role in causing high atherogenic particle concentrations. Unfortunately this pathophysiologic process is not taught in medical schools, residency or even discussed very much at NLA

meetings. It is amazing how many lipidologists simply think LDL size is what explains LDL-C / LDL-P discordance. In essence, TG, at levels never before realized have tremendous importance in our insulin resistant population because they make the liver produce too many TG-rich, large VLDLs which have delayed lipolysis and because of increased CETP activity send their TG to LDLs and HDLs creating CE-poor LDLs and HDLs. There is great potential for LDL-C / LDL-P discordance. The highest LDL particle counts would be seen in those with the most CE-depleted cores – small LDLs that are also TG-rich!

Since TG are so amenable to therapeutic lifestyle interventions, it seems like this should be easy to correct. For those not willing to do that or where lifestyle fails (perhaps in those who have more genetic factors at play): drugs that reduce TG synthesis can be used to help statins achieve LDL-P (apoB, non-HDL-C) goals: fibrates, high dose N-3 fatty acids or high dose niacin.

This is a perfect case illustrating my teachings that LDL-C can be very misleading. So many providers looking at this man's lipid profile would dismiss the CV risk or would accept him to be at NCEP-ATP-III goal. However a lipidologist panics when they see a TG of 217. Note the TG/HDL-C ratio is 4.0. Such ratios > 3.0 in men is associated with significant CV risk and is almost always associated with too many apoB (mostly LDL particles). The non-HDL-C, a better predictor of risk than LDL-C, is also elevated at 154 (TC minus HDL-C) in the second but not the first panel. Non-HDL-C is simply the "poor man's apoB or LDL-P." Desirable according to NCEP ATP-III is well under 130 mg/dL.

As pointed out by Tabs in the article cited at the beginning, the cause of atherogenesis is having too many apoB particles (>90% of which because of its longer half life are LDLs).