Going Beyond Cholesterol

Adding Lp(a), apoB, apoA–1 and hsCRP as an Advanced CV Risk Panel to Improve Mortality Risk Prediction

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Clinical Reference Laboratory

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The standard CRL laboratory panel includes Total Cholesterol (TC or chol) and HDL (more appropriately identified as HDL-C) to assess cardiovascular mortality risk as well as the chol/ HDL-C ratio with any display of triglycerides and low-density lipoprotein (LDL-C) optional. LDL-C can be provided calculated from the Friedewald formula (TC - HDL-C - Trig/5).

However, recent research suggests that other lipid and inflammatory tests have independent or superior value in refining an individual applicant's risk. These tests include **Lp(a), hs-CRP, apoB and apoA-1 and resulting apoB/apoA-1 ratio.** These can be provided as an **Advanced CV (cardiovascular) Risk Panel** for your selected applicants by automatic or manual reflex. Discussion of these tests follow but first a little background including a brief discussion of other tests such as NT-proBNP and HbA1c you should be doing now to better assess CV mortality risk for these applicants before adding the Advanced CV Risk Panel.

Background

Cardiovascular risk assessment for life insurance advanced substantially when the risk associated with hypertension was recognized and risk ratings implemented. Later, the importance of total cholesterol (or LDL-C) and HDL-C levels in CV risk was appreciated and incorporated into risk algorithms. Treatment to reduce LDL-C, especially statins, became common as well with substantial success. However, steadily increasing obesity in the life insurance applicant pool has increased CV risk in a variety of ways but especially through increasing blood sugar generating a HbA1c of 6.0% or higher, the point where mortality risk appears to increase. It was also found that CV mortality is highly associated with, an often unrecognized reduced left ventricular (LV) performance and that the hormone BNP was a highly sensitive and specific predictor of a stressed cardiac ventricle in those with and without any other findings of CV disease. The peptide, NT-proBNP, is substantially easier to transport and equally, if not more, predictive of current (not just some distant future) CV mortality risk.

Optimal testing of life insurance applicants (at least for the higher CV risk at ages of 50+) now includes systolic blood pressure (with optimal SBP being <=120 mm Hg (not <140), HbA1c (directly or reflexed off fructosamine and history), NT-proBNP (which is the best predictor of immediate CV risk), and lipids consisting of total cholesterol (or LDL) and HDL (or chol/HDL ratio). BMI as a measure of obesity assists as well. The advanced CV risk panel will help to adjust

risk upward or downward but would not initially substitute for the existing CV testing noted above.

Advanced CV Risk Panel

Based on more recent research and improved understanding, we can now improve assessment of CV risk by including apoB/apoA-1, Lp(a) and hsCRP as we are missing some important markers of mortality risk and might be over or understating the mortality risk based on the lipid tests in current use.

A major issue in introducing new tests for insurance is to accurately associate test results with expected levels of all-cause mortality. Our existing mortality expectations and any matching underwriting table ratings guidance for lipids are based largely on insurance applicants tested at CRL with mortality usually based on the Soc. Sec. Death Master File. This provided a matching population (insurance applicants) and matching outcome (all-cause mortality) and allowed appropriate division of test values into bands and any needed splits by age and gender. No similar testing (at least insurance testing) has been done for these newer tests so that mortality (relative risk) must be obtained from the literature which typically provides CV morbidity or mortality rather than all-cause mortality. Fortunately, an article by Johannesen in 2020 looked at both CV and all-cause mortality by LDL level using the very large and well-described Copenhagen General Population Study with ages similar to those we anticipate testing.[1] She found the hazard ratio (HR) for all-cause mortality was almost equal to that for CV mortality as LDL varied from 77 to 387 mg/dL At values <77 mg/dL, all-cause mortality increased faster than CV risk as expected because low LDL in older individuals may indicate other conditions such as advanced malignancy. Xiao, in a 2023 article utilizing NHANES data, also found CV mortality and all-cause mortality to change equally for a population stratified by LDL-C/apoB ratio.[2] For the purposes of this paper, CV and all-cause mortality HRs will be assumed to be similar unless both are provided in the source or LDL is <77 mg/dL.

ApoB

We now appreciate that lipids can be transported into arterial walls by multiple lipoprotein particles including very-low-density lipoproteins (VLDL), low-density lipoproteins (LDL) and intermediate-density lipoproteins (IDL) and the best measure of risk is the total number of such particles and not the amount of cholesterol they contain. If we look only at LDL cholesterol or total (or non-HDL) cholesterol we may understate or overstate the CV risk because individual applicants will have different mixes of LDL, IDL, and VLDL particles and have different amounts of cholesterol in each of those particles. Chylomicrons have an apoB48 core which does not penetrate arterial walls so they can be ignored. Because each particle transporting lipids into arterial walls contains one apolipoprotein B (apoB or apoB-100), this is a more accurate measure

and is often used in Europe instead of LDL, but LDL use persists in the U.S. and has been a less expensive test. However, with the obesity-DM epidemic potentially shifting the percentage of each lipoprotein present and with the widespread use of statins, LDL-C may no longer be a sufficiently accurate measure of risk for an individual's risk (rather than comparing CV risks between populations).

Almost every study shows apoB to be a better CV risk and mortality predictor than LDL-C (the worst) or non-HDL-C (better). Although seldom directly compared to apoB, risk prediction for total cholesterol lags that of non-HDL-C only slightly. ApoB is a superior measure of risk because it measures all atheroma-promoting particles (except Lp(a)) and because the amount of cholesterol varies in LDL particles so that LDL-C either under- or over-estimates the total apoB-containing particle count. But LDL persists as the main test utilized to define lipid risk because of a widespread belief that results are close enough and the effort and additional cost of converting to apoB is not worth it. However, in his 2004 article, A. Sniderman, is able to demonstrate exactly how far off the LDL value can be.[3] Using the UK biobank, he shows that even though the correlation coefficient between apoB and LDL is 0.96, for an LDL-C of 130 mg/dL the corresponding range of apoB to capture 95% of these LDLs would be 106.3 to 131.7 mg/dL and this wide range (and varying risk) is apparent at lower and higher LDL-C values as well. When risk of a CV event is considered, the apoB level still has a residual risk with a hazard ratio (HR) of 1.06 per standard deviation after LDL-C is considered and a 1.04 HR residual risk after non-HDL-C is considered. There is no residual risk for LDL or for non-LDL-C after apoB is considered. Two other studies utilizing the Copenhagen General Population Study (Johannesen) and the Framingham Heart Study (Pencina) found similar results for CV event and mortality comparing apoB and LDL.[4,5] LDL may or may not be good enough for some purposes but when you are considering large life policies on individuals there can be substantial difference in risk noted using apoB instead of the less accurate total cholesterol or LDL-C. This difference is why apoB is part of this CRL Advanced CV Risk Panel.

Johannesen also found that apoB has the expected "J" shaped CV mortality with lowest risk around a value of 75 mg/dL with 110% HR at 100 mg/dL, 150% HR at 150 mg/dL and 175%HR at 200 mg/dL.[4] Huang found all-cause mortality to be similar for apoB levels <140 mg/dL but a HR of 175% for those >=140 mg/dL.[6] Lu, also looking at all-cause mortality via Mendelian randomization, found that as compared to risk at 75 mg/dL, risk at 100 mg/dL increased to around 115%, at 125 mg/dL to 130% and at 150 mg/dL to around 150%.[7]

Using apoB for Risk Assessment (also see "using apoB/apoA-1" below)

Rather than try to modify the risk noted for LDL or total cholesterol based on apoB associated risk as shown above, an alternative approach to assessing risk by apoB is to compare apoB levels

with LDL-C levels as a ratio with risk rating reduced if ratio is higher and the risk rating increased if ratio is lower.

One way of doing that is included in an article by Xiao in 2023 where he used NHANES data to look at CV mortality against the LDL-C/apoB ratio.[2] A ratio of 1.25 is the average seen in those without CV death. Using a LDL-C/apoB ratio of 1.2 as neutral, a ratio of 1.0 had a HR of 1.2, ratio of 0.8 had a HR of 1.5 and ratio of 0.6 had a HR of 2x of that based on LDL-C alone. If the LDL-C/apoB ratio is instead >1.2, a ratio of 1.4 has a HR of only 0.9, a ratio of 1.6 has a HR of 0.8, a ratio of 1.7 has a HR of 0.6, and a ratio of 2 has a HR of 0.5 x the LDL-based all-cause mortality risk. To facilitate use of LDL-C/apoB, the LDL-C and LDL-C/apoB ratios will be included in this panel.

An alternative approach is to generate an average LDL based on the measured apoB value and compare that LDLapoB to the LDL measured for that individual. If the LDLapoB is higher than the measured LDL, then risk is higher as there are additional unmeasured risk particles and if lower, then overall CV risk is actually lower than suggested by measured LDL. When presented as a ratio of LDLapoB /LDL, the percentage can be used for lipid risk rating adjustments. Cole published such an apoB to LDL translation in Clinical Chemistry 2003 with LDLapoB-C = 1.38(apoB) - 29 using mg/dL units.[8] This seems a more cumbersome approach than the LDL-C/apoB ratio and will not be further discussed here but could be implemented if client was interested.

ApoA-1 and apoB/apoA-1

There is one apoA-1 particle in each HDL particle along with cholesterol of varying amounts analogous to apoB in non-HDL particles including LDL, VLDL and IDL. There are limited studies of apoA-1 in isolation, but Walldius, utilizing data from the AMORIS long term study published results in 2004 and 2021 (and in other articles) looking at risk of CV mortality relative to apoB, apoA-1, LDL, total cholesterol, HDL and ratios.[9,10] Risk rose steeply as apoA-1 fell in an age-adjusted univariate analysis for both men and for women with an increased risk as apoB increased. When the lowest tertile (116 mg/dL) of apoA-1 was compared to the highest tertile (162 mg/dL), risk decreased by 90% in men and 63% in women. However, it is as yet unclear as to how much better a risk predictor apoA-1 is relative to HDL-C.

There is more data on the apoB/apoA-1 ratio which is analogous to cholesterol/HDL ratio. As compared to TC/HDL-C with an age-adjusted relative risk for each standard deviation (RR/SD) of 1.12 in men and 1.14 in women, Walldius found apoB/apoA-1 to have a RR/SD of 1.42 in men and 1.37 in women which means it is substantially more predictive. In an analysis adjusted for age and for the alternate ratio (apoB/apoA-1 for TC/HDL-C and vice versa) apoB/apoA-1

retained its RR/SD of 1.47 in men and 1.27 in women while TC/HDL-C fell to an RR/SD of .97 in men and 1.08 in women. Almost all the predictive power was owned by apoB/apoA-1.

In the 2021 study, CV mortality was compared by deciles of apoB/apoA-1. Lowest mortality was found at a ratio of .7 (with a .85 RR) with risk increasing up to a 1.0 RR for lower ratios and also increasing for higher ratios- a ratio of .9 had a .9 RR, ratio of 1.0 had a 1.1 RR, ratio of 1.2 had a 1.2 RR, and ratio of 1.5 had a 1.4 RR. When older ages from the WOLF study were also considered, RR rose more quickly reaching 2 for apoB/apoA-1 ratio of 1.3. Nomikos, in 2015 using the ATTICA study, found that all-cause mortality risk increased by .0.4% per 1 mg/dL increase in apoB and decreased by 0.4% per 1 mg/dL increase of apoA-1 with risk increased by 56% by a 1 unit increase in apoB/apoA-1. The apoB/apoA-1 had the best performance (of lipid measures) in reclassifying status (dead or alive) based on all other non-lipid measurements and history.[11] Zhang published a review of apo studies in 2024 finding that the association between all-cause mortality and apoB/apoA-1 to be strong with more inconsistent support for apoA-1 or apoB alone.[12] Heterogeneity of results was high making a meta-analysis complex.

Using apoB/apoA-1 for Risk Assessment

This ratio is analogous to TC/HDL-C (or more correctly to non-HDL-C/HDL-C) but more accurately predicts CV and all-cause mortality because it measures particle number rather than mass of particles which may vary. CRL, using life insurance applicants, as well as other authors using other cohorts have found TC/HDL-C and non-HDL-c/HDL-C to be the best and simplest risk predictor for all-cause mortality related to lipid levels obviating the need to look at multiple values. For our current purposes in using this advanced CV risk panel, we are trying to supplement currently used lipid testing in modifying underwriting risk class up or down. This will typically impact preferred risk classes and rarely a standard risk class unless other risks are present as well.

ApoB/apoA-1 ratios from 0.6 to 0.8 in women and 0.6 to 0.9 in men are associated with the lowest risk which may be 10% less than the next band. Ratios above this up to 1.1 have an average risk and ratios above 1.1 have an increasing risk. Ratios of 1.5 suggest an all-cause mortality that may exceed 125% and ratios 1.6+ are likely associated with risk of 150% or higher. Ratios <0.6 may suggest the presence of very low TC or very high HDL values also needing evaluation (for ill health, poor diet, heavy alcohol, etc).

Lp(a)

Another recent advancement in lipid research is the appreciation of the risk associated with the Lp(a) lipid particle which consists of an apo(a) attached to an apoB and associated cholesterol. The Lp(a) level is metabolically distinct from other lipids and is almost entirely genetically

determined. That risk is additive to the risk associated with other apoB containing lipids and is not currently adequately measured by either apoB or cholesterol testing nor can it be treated by statins or other drugs aimed at LDL. The Lp(a) level does not correlate with any other measures of CV risk and must be measured separately in either mg/dL (total mass) as has been common in the U.S. or in nmol/L (number of particles) which is a better risk measure as Lp(a) particle size can vary dramatically). Nmol/L is used everywhere else in the world. Recently, Roche Diagnostics received the first FDA-cleared test in the US to measure Lp(a) in Nanomoles per liter (nmol/L). Availability is expected by September, in the meantime CRL will continue to run the FDA-cleared Lp(a) mg/dL assay. Previously, Lp(a) could be measured but not treated if elevated and as such was of less interest clinically, but now highly effective Rx will soon be available as well. An in-depth review of Lp(a) and its use for risk assessment is available in our CRL Lp(a) whitepaper.

In addition to studies in the whitepaper, a 2024 paper in JAMA by Small utilized 3 different datasets including 2 from TIMI trials and the UK biobank.[13] The investigators split the cohorts into those with hs-CRP <2 mg/L and those higher and looked at CV events (MACE) and CV mortality by 50 nmol/L bands of Lp(a) finding Lp(a) risk to be independent of hs-CRP and increasing MACE by 5% and CV mortality by 1-2%. Splitting the cohorts by Lp(a) at 125 nmol/L and hs-CRP at 2 mg/L resulted in similar additive increased risk of MI for higher CRP, higher Lp(a) and when both were higher.

Using Lp(a) for Risk Assessment

Reviewing the data in the CRL Lp(a) whitepaper and newer data and looking at results in nmol/L (particle count) rather than mg/dL (mass including cholesterol), the onset of additional risk appears to begin at approximately 125 nmol/L which is near the 80th percentile of distribution. Assuming 50 mg/dL is approximately equivalent to 125 nmol/L, each additional 125 nmol/L might increase all-cause mortality by 5-10%. A 25% increase (or one mortality table) would require Lp(a) values >400 nmol/L impacting <0.05% of applicants. All mortality studies reviewed suggest there is no reduced risk (preferred risk) pool associated with Lp(a) but assuming avoidance of even small amounts of excess risk is desired for a preferred class, values >125 (approximately 8oth percentile) would likely be associated with a small increased risk. If other CV risk factors are present, such levels might lead to treatment when (probably soon) FDA licensed Lp(a) treatment becomes available and would have a risk more consistent with a standard class.

hsCRP

The third test is high sensitivity C-reactive protein (hsCRP or just CRP). This is not actually a new test and debate has raged as to whether it had independent predictive value for CV and all-cause mortality. Performing it accurately utilizing insurance applicant samples has also been an issue. More recently both issues appear to have been resolved. CRL can currently produce

accurate reproducible hsCRP results and more recent research documents its value in risk prediction.

hsCRP values are higher in women but the sex difference appears to relate to estrogen levels so that values even out for post-menopausal women. For ages 50-69, roughly 40% of US adults have values <2.0 mg/L, 15% have values 2.0-2.9, 34% have values 3.0-9.9 and the remaining 11% have higher values.[14] More black persons had values ≥10 ng/L than white persons.

The most notable study on hsCRP in CV risk is the one from NEJM by P. Ridker utilizing the Women's Health Study data which followed professional women for over 30 years finding hsCRP, lp(a) and LDL all independently predictive of occurrence of the first CV event.[15] CRP had the greatest impact with an adjusted hazard ratio (HR) of 1.14 per quintile, LDL with 1.09 per quintile and Lp(a) with an adjusted HR of 1.07 per quintile but essentially all of that increased Lp(a) risk (HR of 1.33) appeared in the highest quintile of Lp(a) unlike hs-CRL and LDL where each quintile had increasing risk.

Another study of Lp(a) and CRP was conducted by Small, and results are included above under Lp(a).[13]

Ridker was also the first author on the report in 2008 of the Jupiter trial treating healthy men and women with normal LDL levels (<130 mg/dL) but higher levels of hsCRP levels of ≥2.0 mg/L which was stopped early due to such impressive reduction in CV events and mortality (20%) with an associated reduction in LDL of 50% and CRP of 37%.[16] CV event reduction was estimated to be twice what would have been expected based on LDL reduction alone.[10]

Youssuf in 2013 provided a more nuanced review of hsCRP. He points out limitations in the JUPITER study, the association between higher hsCRP levels and other CV risk factors including hypertension, metabolic syndrome, age and estrogen (premenopausal women) and the inconsistent reclassification of risk class based on addition of hsCRP testing.[17] He also discusses that CRP levels are genome dependent and vary between ethnic groups without evidence that higher values are associated with more CV disease across populations.

Using hsCRP for Risk Assessment

HsCRP values in the age50+ group <2.0 mg/L are favorable and if other CV risk factors make a preferred underwriting class offer borderline, an hsCRP value of <2.0 mg/L would likely negate much of the risk. Values >10 mg/L will occur in <10% of applicants and if are unfavorable and if other risk factors are present, possibly increasing risk beyond a typical standard underwriting

class. Values between 2.0 and 10 mg/L have progressively increased risk but some of that risk may already be accounted for obesity, increased HbA1c, hypertension, etc.

Modifying Original Risk Assessment Based on the CRL Advanced CV Risk Panel Results

These 4 tests, when applied to a pool of applicants with at least moderate risk of CV events potentially leading to death within a 10 year time frame (= age 50+ or equivalent), can be helpful in discovering a small number of applicants with unexpectedly high risk who would be moved out of a standard risk class and, for a larger number of applicants, in refining preferred and standard class eligibility when other test values or histories are borderline. This panel may also provide applicants found to have higher risk with hard-to-get but desirable information on how to improve their cardiovascular health and longevity.

The advanced panel will provide apoB in mg/dL, apoA-1 in mg/dL and the resulting apoB/apoA-1 ratio, Lp(a) in nmol/L, hsCRP in mg/L, LDL in mg/dL calculated from Friedewald formula and LDL-C/apoB. With guidance provided above, the apoB/apoA-1, Lp(a) and hsCRP test results are the critical values to use in modifying the original underwriting assessment. The "Using *test* for risk assessment" sections for each panel component provide the specific information you can utilize in creating your own underwriting guidelines for the Advanced CV Risk Panel and we would be happy to discuss those translations further with you.

When to Use the Advanced CV Risk Panel

Because of additional testing cost and the modest overall risk reduction, even though impact on individual risk may be substantial, it is suggested that this testing initially be applied to those applying for higher face amounts who because of age (50+) or other risk factors have a reasonable risk of having at-risk atherosclerotic lesions which could lead to early mortality risk. Such individuals may also appreciate the test results which generally have limited availability at high cost. A generic guide to applicants to interpret these results is available from CRL. The panel may also prove useful and be manually ordered from CRL when the appropriate underwriting class based on CV risk is uncertain at any age.

References

1. Johannesen CD, Langsted A, Mortensen MB, Nordestgaard BG. Assoc. between LDL and all-cause and cause-specific mortality in Denmark. BMJ 2020;371:m4266. http://dx.doi.org/10.1136/bmj.m4266
2. Xiao L, Zhang K, Fang W, et al. The LDL-C/apoB ratio predicts CV and all-cause mortality in the general population. *Lipids in Hlth and Dis* 2023;22:104. https://doi.org/10.1186/s12944-023-01869-1
3. Sniderman AD, Dufresne L, Pencina KM, et al. Discordance among apoB, non-HDL cholesterol and triglycerides *European Heart Journal* 2024:45:2410-2418. https://doi.org/10.1093/eurheartj/ehae258
4. Johannesen CD, Mortensen MB, Langsted A, Nordestgaard BG. Apolipoprotein B and non-HDL cholesterol better reflect residual risk than LDL-C in statin-treated patients. *JACC* 2021;77:1439-50. https://doi.org/10.1016/j.jacc.2021.01.027
5. Pencina MJ, D'Agostino RB, Zdrojewski T, et al. Apolipoprotein B improves risk assessment of future coronary heart disease in the Framingham heart study beyond LDL-C and non-HDL-C. *Eur J Prev Cardiol* 2015;22:1321-27. https://doi.org/10.1177/2047487315569411
6. Huang Y, Chen S, Huachun P, Yang S, Cheng W. Relationship between serum apolipoprotein B and risk of all-cause and cardiovascular disease mortality in individuals with hypertension. *BMC Cardiovasc Dis* 2024;24:273. https://doi.org/10.1186/s12872-024-03949-1
7. Lu J, Wang Z, Zhang J, et al. Causal association of blood lipids with all-cause and cause-specific mortality: a Mendelian randomization study. *J Lipid Res* 2024;65:100528. https://doi.org/10.1016/j.jlr.2024.100528
8. Cole J, Otvos JD, Remaley AT. A translational tool to facilitate use of apoB for clinical decision making. *Clin Chem* 2023;69:41-47. https://academic.oup.com/clinchem/article/69/1/41/6821418
9. Walldius G, Jungner I, Are HA, et al. The apoB/apoA-1 Ratio is better than cholesterol ratios to predict coronary risk. *Clin Chem Lab Med* 2004;42:1355-63. https://doi.org/10.1515/CCLM.2004.254
10. Walldius G, de Faire U, Alfredsson L, et al. Long-term risk of a major CV event by apoB, apoA-1 and apoB/apoA-1 ratio. *Plos Medicine* 2021. https://doi.org/10.1371/journal.pmed.1003853
11. Nomikos T, Panagiotakos D, Georgousopoulou E, et al. Hierarchical modelling of blood lipids and 10-year all-cause mortality. *Lipids in Hlth and Dis* 2015;14. https://doi.org/10.1186/s12944-015-0101-7

12. Zhang J, Zinru S, Zhi L, et al. Association of apolipoprotein levels with all-cause and CV mortality. *Eur J of Prev Cardiol* 2024;31:1183-94. https://doi.org/10.1093/eurjpc/zwae080
13. Small AM, Ashley P, Melloni GE, et al. Lipoprotein(a), C-reactive protein and cardiovascular risk in primary and secondary prevention populations. *JAMA Cardiology* 2024;9:385-391. https://doi.org/10.1001/jamacardio.2023.5605
14. Woloshin S, Schwartz LA. Distribution of C-reactive protein values in the United States. *NEJM* 2005;352:1611-13. https://doi.org/10.1056/NEJM200504143521525
15. Ridker PM, Moorthy MV, Cook NR, et al. Inflammation, cholesterol, lipoprotein(a) and 30-yr cardiovascular outcomes in women. *NEJM* 2024;391:2087-97. https://doi.org/10.1056/NEJMoa2405182
16. Ridker PM, Danielson E, Fonseca FA, et al. Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. *NEJM* 2008;359:2195-207. https://doi.org/10.1056/NEJMoa0807646
17. Yousuf O, Bibhu DM, Martin SS, et al. *JACC* 2008;62:397-408. http://dx.doi.org/10.1016/j.jacc.2013.05.016