疾病研究 :血浆靶向脂质组学轮廓谱研究提高二型糖尿病病人中患心血管疾病的传统风险因子的预测能力

研究对象：人
分析检测平台：LC ESI-MS/MS
期刊：Circulation
影响因子：17.047
发表时间：2016

摘要：

Background: Clinical lipid measurements do not show the full complexity of the altered lipid metabolism associated with diabetes or cardiovascular disease. Lipidomics enables the assessment of hundreds of lipid species as potential markers for disease risk. Methods: Plasma lipid species (310) were measured by a targeted lipidomic analysis with liquid chromatography electrospray ionisation-tandem mass spectrometry on a case-cohort (n=3,779) subset from the ADVANCE (Action in Diabetes and Vascular disease: preterAx and diamicroN- MR Controlled Evaluation) trial. The-case cohort was 61% male with a mean age of 67. All participants had type 2 diabetes mellitus with one or more additional cardiovascular risk factors and 35% had a history of macrovascular disease. Weighted Cox regression was used to identify lipid species associated with future cardiovascular events (non-fatal myocardial infarction, non- fatal stroke and cardiovascular death) and cardiovascular death during a five year follow-up period. Multivariable models combining traditional risk factors with lipid species were optimized using the Akaike information criteria. C-statistics and net reclassification indices (NRI) were calculated within a five-fold cross validation framework. Results: Sphingolipids, phospholipids (including lyso- and ether- species), cholesteryl esters, and glycerolipids were associated with future cardiovascular events and cardiovascular death. The addition of 7 lipid species to a base model (14 traditional risk factors and medications) to predict cardiovascular events increased the C statistic from 0.680 (95% confidence interval [CI], 0.678 - 0.682) to 0.700 (95% CI, 0.698 - 0.702; P<0.0001) with a corresponding continuous NRI of 0.227 (95% CI, 0.219 - 0.235). The prediction of cardiovascular death was improved with the incorporation of 4 lipid species into the base model, showing an increase in the C statistic from 0.740 (95% CI, 0.738 - 0.742) to 0.760 (95% CI, 0.757 - 0.762; P<0.0001) and a continuous net reclassification index of 0.328 (95% CI, 0.317 - 0.339). The results were validated in a subcohort with type 2 diabetes mellitus (n=511) from the LIPID trial (Long-Term Intervention With Pravastatin in Ischemic Disease). Conclusions: The improvement in the prediction of cardiovascular events, above traditional risk

factors, demonstrates the potential of plasma lipid species as biomarkers for cardiovascular risk stratification in diabetes mellitus.

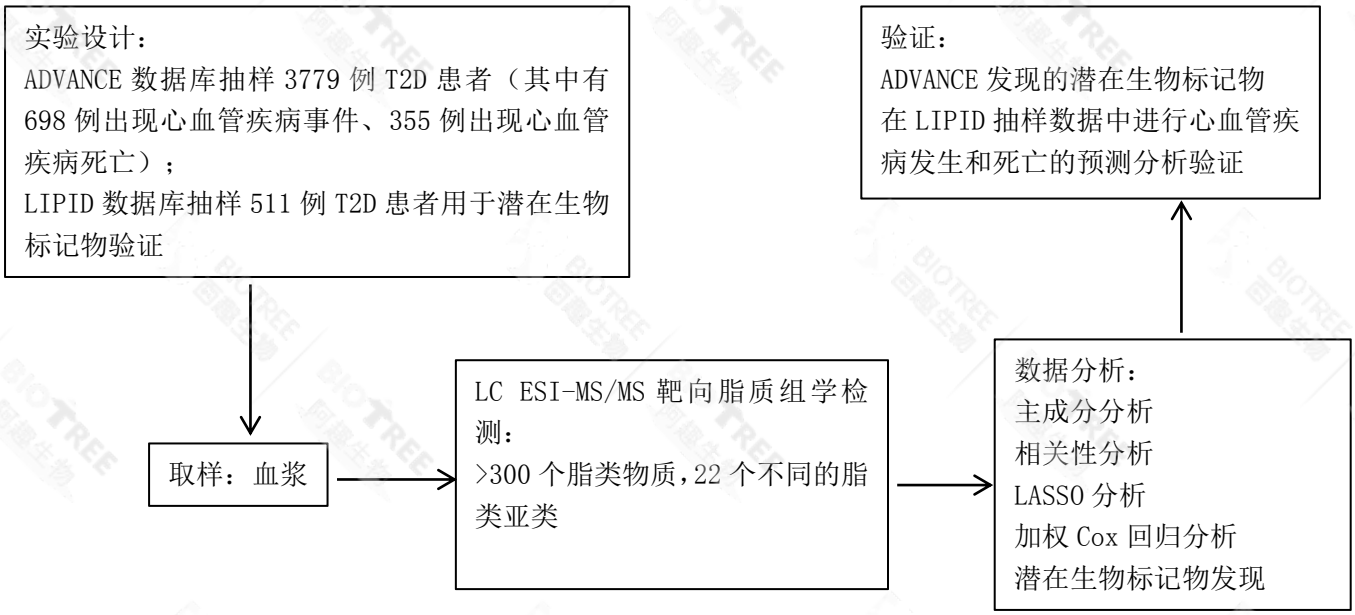
Keywords: lipids, biomarker, mass spectrometry, cardiovascular outcomes, diabetes mellitus.

一、研究背景：

二型糖尿病（T2D）是一种全球各国经济和健康负担日益增加的疾病，动脉粥样硬化性心血管疾病（CVD）是 T2D 并发症中致死率最高的并发症，评估和调控 CVD 的发生是 T2D 治疗中的一个重点内容。虽然已经建立了 CVD 风险打分模型，但是弗雷明汉风险评分低估了 T2D 患者患 CVD 的风险，而英国糖尿病风险评分高估了 T2D 患者患 CVD 的风险。

传统的脂类物质标记物（总胆固醇、低密度脂蛋白胆固醇、三酰甘油和高密度脂蛋白胆固醇）被用于风险评分，但是这些有限的检测都无法解释 T2D 及其相关心血管疾病复杂的脂类变化。脂质组学技术的发展以及血浆中越来越多的脂类物质被发现与 T2D 和心血管疾病密切相关，本文拟通过使用脂质组学的方法研究和评估 T2D 人群患心血管疾病的风险，希望能够找到较好预测 5 年后是否会患心血管疾病的脂类物质。

二、方法流程：



三、研究结果与讨论：

1 临床信息特征：

1）基于各项临床指标信息，发现在后续跟踪中发生心血管疾病病发事件或者心血管疾病病发死亡的人群具有更高的HbA1c和SBP、更长时间的糖尿病史、更可能有心血管疾病史、普遍锻炼较少、更低的HDLC和eGFR。

Variable*	All (n=3779)	Cardiovascular events (n=698)	No cardiovascular events (n=3,081)	*p*-value†	Cardiovascular death (n=355)	No cardiovascular death (n=3,424)	*p*-value†
Continuous variables, median (1st, 3rd quartile)							
Age (years)	67 (62, 72)	70 (65, 74)	67 (61, 71)	**<0.001**	71 (66, 75)	67 (61, 71)	**<0.001**
Body mass index (kg/m^2)	29.4 (26.4, 32.8)	28.7 (26.1, 32.5)	29.4 (26.6, 32.9)	**0.030**	28.7 (26.1, 32.4)	29.4 (26.5, 32.9)	0.080
HbA1c (%)	7.2 (6.5, 8.1)	7.3 (6.5, 8.4)	7.1 (6.4, 8.1)	**<0.001**	7.5 (6.6, 8.5)	7.1 (6.4, 8.1)	**<0.001**
Glucose (mmol/L)	7.9 (6.6, 9.8)	8.1 (6.6, 10.1)	7.9 (6.6, 9.7)	0.163	8.2 (6.5, 10.3)	7.9 (6.6, 9.7)	0.321
Triglycerides (mmol/L)	1.70 (1.20, 2.35)	1.62 (1.20, 2.32)	1.70 (1.20, 2.36)	0.431	1.60 (1.20, 2.32)	1.70 (1.20, 2.36)	0.397
LDL cholesterol (mmol/L)	3.00 (2.35, 3.70)	3.00 (2.40, 3.80)	2.99 (2.34, 3.70)	0.479	3.05 (2.40, 3.80)	2.99 (2.33, 3.70)	0.198
Total cholesterol (mmol/L)	5.00 (4.30, 5.81)	5.00 (4.30, 5.80)	5.00 (4.31, 5.83)	0.278	5.04 (4.30, 5.88)	5.00 (4.30, 5.81)	0.819
HDL cholesterol (mmol/L)	1.20 (1.00, 1.40)	1.10 (0.96, 1.30)	1.20 (1.00, 1.40)	**<0.001**	1.10 (1.00, 1.33)	1.20 (1.00, 1.40)	**0.011**
Systolic blood pressure (mmHg)	146 (133, 160)	150 (135, 166)	145 (132, 160)	**<0.001**	149 (135, 165)	146 (133, 160)	**0.006**
Diastolic blood pressure (mmHg)	81 (74, 89)	82 (74, 89)	81 (74, 89)	0.951	81 (73, 89)	81 (74, 89)	0.235
eGFR (mL/min/1.73m^2)	71 (60, 85)	68 (55, 81)	72 (61, 86)	**<0.001**	67 (52, 79)	72 (61, 85)	**<0.001**
Diabetes duration (years)	6.0 (3.0, 11.0)	8.0 (4.0, 13.0)	6.0 (3.0, 11.0)	**<0.001**	9.0 (4.0, 15.0)	6.0 (3.0, 11.0)	**<0.001**
C-reactive protein (mg/L)	1.83 (0.87, 4.09)	2.02 (0.93, 4.41)	1.79 (0.86, 4.05)	**0.026**	2.05 (1.01, 4.55)	1.80 (0.86, 4.05)	**0.027**
Dichotomous variables, n (%)							
Sex (male)	2308 (61.1%)	483 (69.2%)	1825 (59.2%)	**<0.001**	240 (67.6%)	2068 (60.4%)	**0.038**
Alcohol drinker	1557 (41.2%)	272 (39.0%)	1285 (41.7%)	0.309	125 (35.2%)	1432 (41.8%)	0.065
Smoker	565 (15.0%)	100 (14.3%)	465 (15.1%)	0.637	48 (13.5%)	517 (15.1%)	0.464
History of macrovascular disease	1321 (35.0%)	343 (49.1%)	978 (31.7%)	**<0.001**	187 (52.7%)	1134 (33.1%)	**<0.001**
History of heart failure	175 (4.6%)	61 (8.7%)	114 (3.7%)	**<0.001**	45 (12.7%)	130 (3.8%)	**<0.001**
Use of antihypertensive medication	3022 (80.0%)	607 (87.0%)	2415 (78.4%)	**0.022**	322 (90.7%)	2700 (78.9%)	**0.018**
Use of lipid-lowering medication	1674 (44.3%)	295 (42.3%)	1379 (44.8%)	0.371	140 (39.4%)	1534 (44.8%)	0.148
Use of antiplatelet medication	1869 (49.5%)	411 (58.9%)	1458 (47.3%)	**<0.001**	220 (62.0%)	1649 (48.2%)	**<0.001**
Antihypertensive treatment arm	1850 (49.0%)	332 (47.6%)	1518 (49.3%)	0.561	154 (43.4%)	1696 (49.5%)	0.115
Glucose control arm	1890 (50.0%)	340 (48.7%)	1550 (50.3%)	0.590	166 (46.8%)	1724 (50.4%)	0.363
Moderate or vigorous exercise‡	1822 (48.2%)	285 (40.8%)	1537 (49.9%)	**0.002**	134 (37.7%)	1688 (49.3%)	**0.003**

图 1 临床信息特征

2 脂类物质与心血管疾病发生风险的关联分析：

1）22 类脂类亚类中有 3 类脂类亚类与心血管疾病发生风险具有显著关联；有 2 类脂类亚类与心血管疾病死亡风险具有显著关联；

2）32个脂类物质与未来心血管疾病发病和死亡具有显著关联：其中27个脂类物质与未来心血管疾病发病呈现正相关作用， 5个脂类物质与心血管疾病发生呈现负相关作用；而有31个脂类物质与心血管疾病死亡具有正相关作用，1个脂类物质与心血管疾病死亡呈现负相关作用。

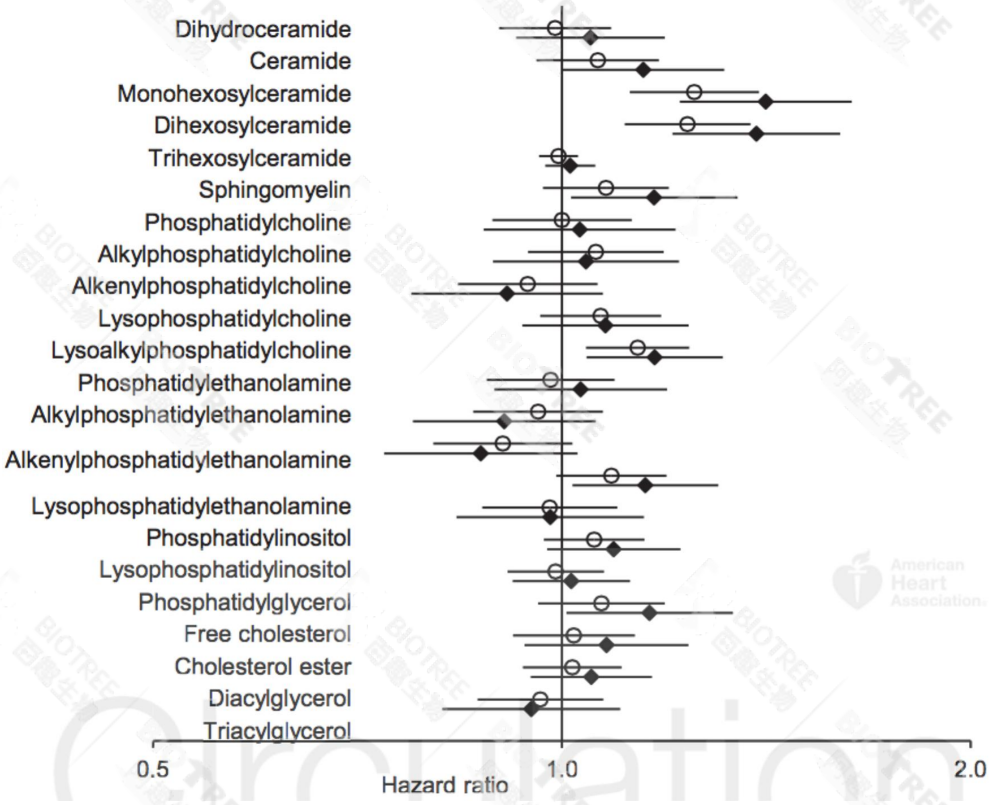


图 2 脂类亚类与未来心血管疾病发生的关联分析

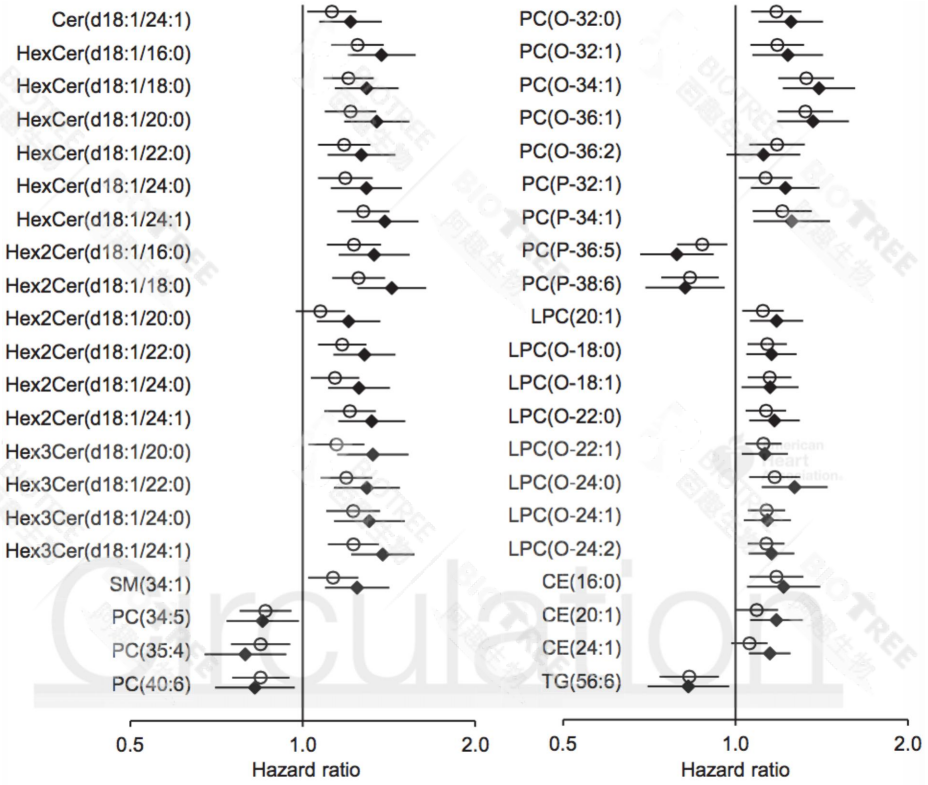


图 3 脂类物质与未来心血管疾病发生的关联分析

3 敏感性分析：

1） 回归分析表明降脂药的使用或者总胆固醇对于脂类物质用于心血管疾病发生和死亡的风险判断没有显著性影响。

4 主成分分析和相关性分析：

1） 整个脂类代谢物轮廓谱的主成分分析的 PC1、PC2、PC3 分别能够解释 24.6%、11.1%、8.7%的变量；

2） 42 个与心血管疾病发生相关的脂类物质间具有复杂的相关性。

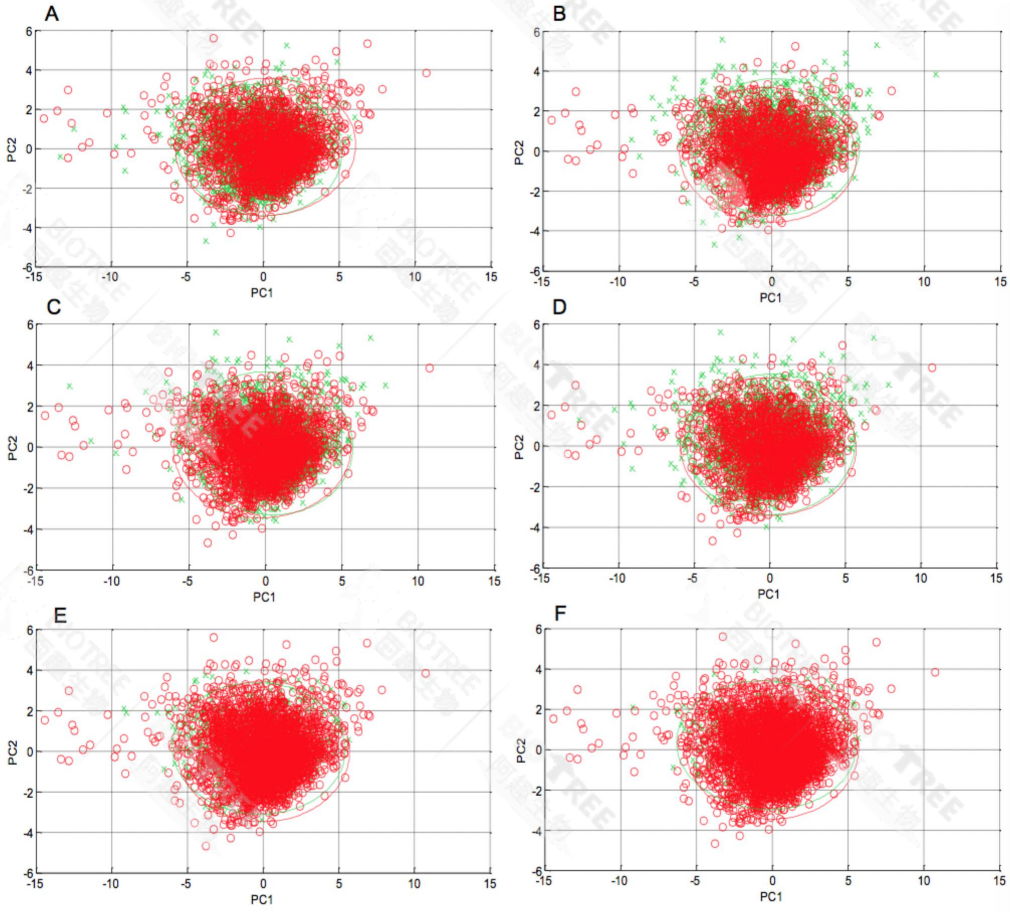


图 4 血浆脂类物质的主成分分析得分图

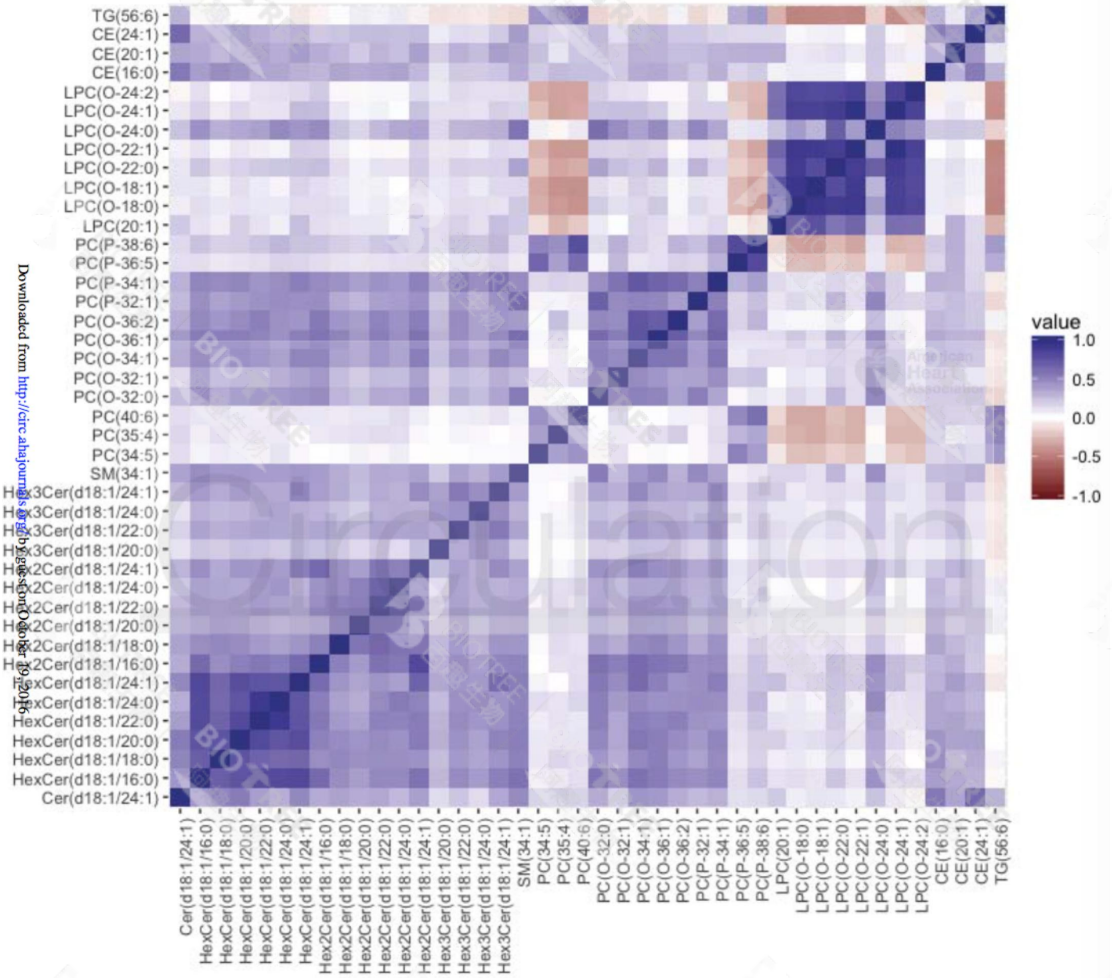


图 5 与心血管疾病发生相关的脂类物质的相关性分析

5 未来的心血管疾病发生和死亡的预测分析：

1） 7 个脂类物质能够提高传统风险因子对心血管疾病发生的预测能力；

2） 4 个脂类物质能够提高传统风险因子对心血管疾病死亡的预测能力，。

6 潜在生物标记物在 LIPID 病人数据库抽样验证：

1） 加入心血管疾病发生的预测模型中的 7 个脂类物质里，在 ADVANCE 数据库抽样样本中 PC(35:4)、PE(O-36:4)与疾病发生呈现负相关作用，而在 LIPID 数据库抽样样本中无显著性作用；此外，在心血管疾病死亡的预测模型中，PC(O-36:1)与疾病死亡呈现负相关作用，而在 LIPID 数据库抽样样本中无显著性作用；

2） 7 个脂类物质能够提高 LIPID 数据库抽样样本中传统风险因子对心血管疾病发生的预测能力；

3） 4 个脂类物质能够提高 LIPID 数据库抽样样本中传统风险因子对心血管疾病死亡的预测能力。

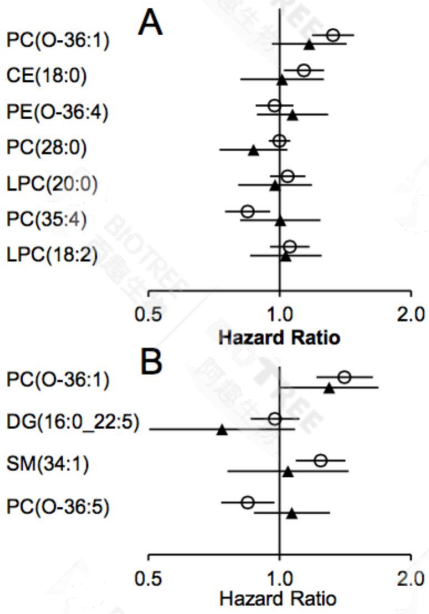


图 6 ADVANCE 与 LIPID 病人数据库与心血管疾病发生相关的脂类物质的相关性分析

四、亮点和展望

1 发现血浆里有 32 个脂类与心血管疾病的发生和死亡具有密切关联

2 将现有的临床诊断风险因子与新发现的 4-7 个脂类物质结合起来，能够提高预测 T2D 患者未来患心血管疾病风险的能力

A 脂类物质的检测数量在本研究中受限，需要进一步研究并提高脂类物质的检测上限

B 不同脂类物质和临床指标间的协同作用对心血管疾病的预测具有重要作用，这对于回归分析是下一项挑战

C ADVANCE 数据库里大部分都是白种人的信息，因此可能不适用于所有的人种，此外，LIPID 数据库的抽样样本较小且部分临床信息与 ADVANCE 数据库不匹配，因此，后续需要加大不同人种和样本进行深入研究

五、文献信息

Alshehry ZH, et al. Plasma Lipidomic Profiles Improve on Traditional Risk Factors for the Prediction of Cardiovascular Events in Type 2 Diabetes Mellitus. Circulation. 2016 Nov 22;134(21):1637-1650.