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Apabetalone (RVX-208) Lowers Major Adverse Cardiovascular Events (MACE) in Diabetes Mellitus by Affecting Complement Pathway and Microbiome Activity

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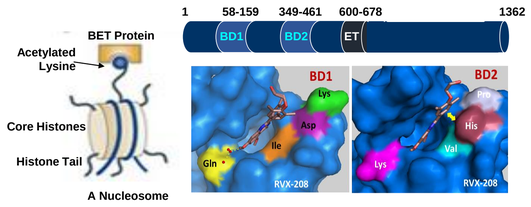
Abstract

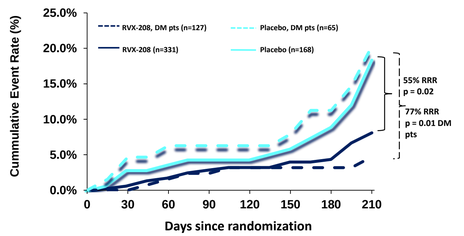
In phase 2b trials lasting 6 months, patients (n=499) given 200 mg/d of apabetalone had a 55% relative risk reduction in MACE that was lowered further in diabetes mellitus (DM). These findings underpin our interest to explore potential mechanism(s) behind MACE reductions arising from RVX-208, a selective BET inhibitor (BETi) that acts via displacing acetylated-lysine moieties in histones from the 2nd ligand domain in bromodomain extra-terminal (BET) proteins. Previous studies showed RVX-208 may lower CVD risks by affecting inflammation, metabolism, coagulation and complement. Studies here examine how RVX-208 may block actions of a suspected culprit in atherosclerosis, trimethyl amine oxide (TMAO) derived from microbiome activity. Huh-7 hepatoma cells or primary hepatocytes (PH) were examined using RT-PCR and western-blots. In high glucose (25.6 mM) Huh-7 cells exposed to 1-100 uM TMAO acted within 6-48 hrs to induce mRNA encoding Mannose Binding Lectin-2 (MBL2) and Complement-3 (C3) members of the complement pathway by 40 and 20%, respectively in a time and dose dependent manner. RVX-208 blocked TMAO induction of MBL2 and C3 in these cells. However, TMAO had no effect on complement C5. In contrast, all 3 genes in euglycemia (5.6 mM) was not induced by TMAO but RVX-208 still lowered their expression. The microbiome transforms dietary phospholipids to TMA the substrate for hepatic flavin mono-oxygenase-3 (FMO3) converting it to TMAO. In PH, RVX-208 acted within 24 hrs to repress FMO3 mRNA and protein levels by 40 and 30%, respectively. In summary, hepatoma cells and hepatocytes cultured in high or euglycemic media reflect diabetic state or not, respectively. TMAO induces complement MBL2 and C3 in cells exposed to hyperglycemia while RVX-208 blocks this induction. Additionally, BETi lowers FMO3 mRNA and protein. Thus RVX-208 blocks both detrimental effects of TMAO and its production as potential mechanisms for how apabetalone lowers MACE in DM.

Background

1.) RVX-208 selectively inhibits BD2 in BET proteins

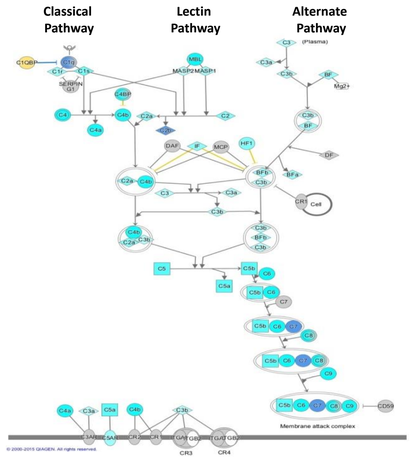


2.) RVX-208 lowers MACE, phase 2b post-hoc analysis



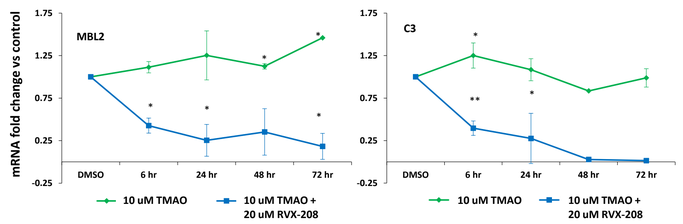
Source: RVX data on file – ASSURE and SUSTAIN Safety Population. Log-Rank test for between group comparison. RRR: relative risk reduction.

Micro-array Gene Sets	Donor 1 Condition 1 NES	Donor 2 Condition 1 NES	Donor 2 Condition 2 NES
REACTOME COMPLEMENT CASCADE	-2.33	-1.92	-2.07
REACTOME FORMATION OF FIBRIN CLOT CLOTTING CASCADE	-2.00	-1.55	-1.75
ACUTE PHASE RESPONSE SIGNALING	-1.76	-1.84	-1.64
KEGG COMPLEMENT AND COAGULATION CASCADES	-2.38	-1.76	-2.05

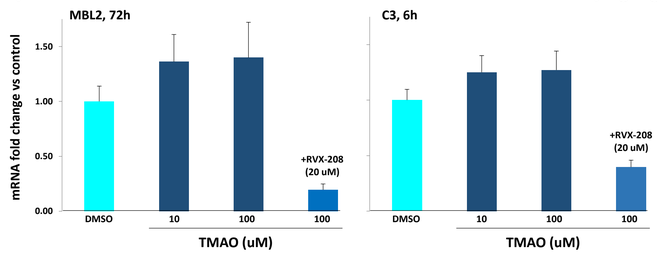


Results

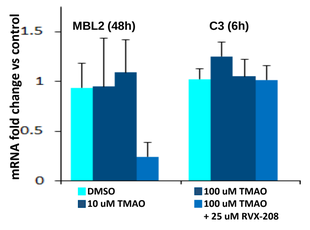
A.) TMAO induces MBL2 and C3 in Huh-7 (high glucose) time course.



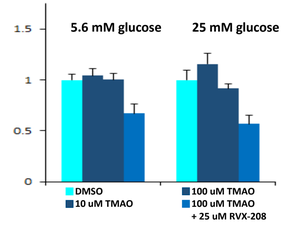
B.) RVX-208 blocks TMAO induction of MBL2 & C3 in Huh-7 (high glucose).



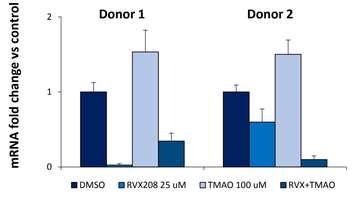
C.) TMAO has no affect on MBL2 or C3 in Huh-7 (euglycemia).



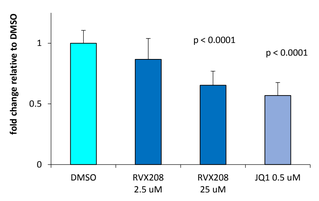
D.) TMAO has no affect on C5 in Huh-7 (euglycemia or high glucose, 48h).



E.) RVX-208 blocks TMAO induction of MBL2 in PHH (high glucose).



F.) FMO3 mRNA is suppressed by BETi using RVX-208/JQ-1



Summary

1.) Apabetalone (RVX-208) lowers MACE in post-hoc analysis of phase 2b studies.

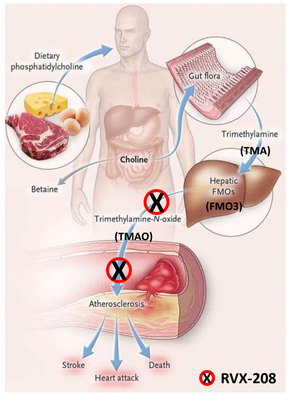
2.) Benefits of RVX-208, a selective BETi, is multifactorial including the complement pathway.

3.) In liver cells, TMAO a dietary by-product suspected to play a role in CVD risks, induces complement MBL2 & C3 but not C5.

4.) TMAO effects are reversed by RVX-208 & provides a potential mechanism for normalizing complement in CVD to lower MACE.

5.) RVX-208 appears to lower FMO3 mRNA, the enzyme responsible for creating TMAO from dietary phospholipids.

TMAO arising from hepatic FMO3 is pro-atherosclerotic & -thrombotic.



Source: Tang, WHW, N. Engl. J. Med. 2013, 368:1575-1584.

Disclosures (All authors are paid employees of Resverlogix Corp.)

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