

# Overcoming the Safety Challenges of Aldose Reductase Inhibition: Development of AT-001 for Diabetic Cardiomyopathy

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## Abstract

Diabetic Cardiomyopathy (DbCM) leading to overt heart failure is a common sequelae of both Type 1 and Type 2 Diabetes. Previous attempts to develop treatments for DbCM through inhibition of the enzyme Aldose Reductase (AR) were unsuccessful, due to low AR binding affinity and poor specificity. Off-target inhibition of Aldehyde Reductase (AldR), an enzyme critical for detoxification of aldehydes in the liver and normal hepatocyte physiology, led to common liver-related safety and tolerability issues with first generation ARI compounds. We report on the selectivity and specificity of AT-001, a novel small molecule ARI with optimized affinity and specificity for AR and minimal to zero off-target AldR activity.

**Methods and Results:** AT-001 was evaluated for AR binding affinity as compared to zopolrestat, a first-generation ARI. AT-001 demonstrated a >300-fold greater affinity for AR versus zopolrestat (IC<sub>50</sub> of 30 pmol for AT-001 and ~10 nmol for zopolrestat). In contrast to zopolrestat, AT-001 showed no inhibition of AldR at 50x and 100x EC<sub>50</sub> levels in assay medium, while zopolrestat inhibited Aldehyde Reductase by 50% and 60% respectively (spec activity of 2.3 and 1.9 mmol NADPH/min/ mg protein). In addition to these analyses, AT-001 was evaluated in a standard off-target receptor binding analysis of 87 substrates (13 enzyme, 74 binding assays) to characterize pharmacological specificity and selectivity. No significant off-target results (defined as ≥50% inhibitory activity) were observed in this panel.

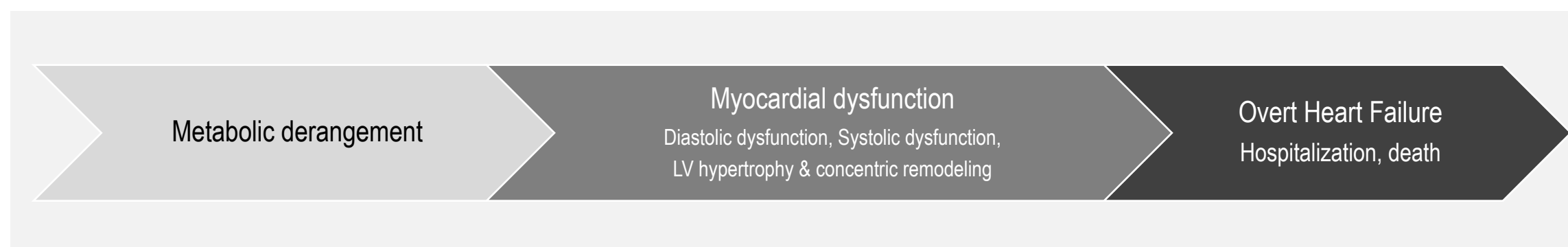
**Conclusion:** The unique structure and activity of AT-001 provide selectivity for Aldose Reductase and lack of off-target effects. The in-vitro safety of this agent together with the positive safety data from the phase 1/2 program, supports the ongoing pivotal study in DbCM.

## Definition of Diabetic Cardiomyopathy (DbCM)<sup>1</sup>

- Abnormal cardiac structure and/or performance

- Resulting from diabetes-associated metabolic alterations
- In the absence of coronary artery disease (CAD) as well as hypertensive, valvular or congenital heart disorder

- Progresses to overt heart failure (HF)<sup>2,3</sup>



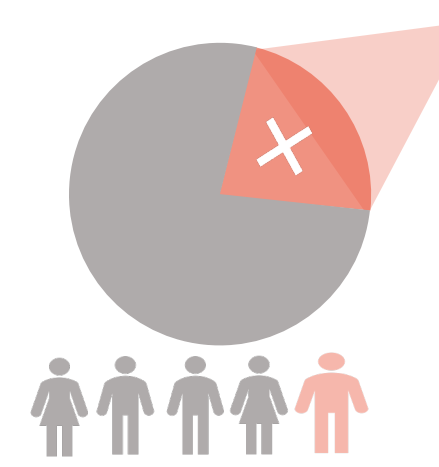
1. Ryden L. Eur Heart J. 2013; 34:3035-3087. 2. Jia G, et al. Circ Res. 2018;122:624-638. 3. Borghetti et al. Frontiers in physiology 2018;9:1514

## Diabetic Cardiomyopathy as a Form of Stage B Heart Failure<sup>1-4</sup>

Stage of Heart Failure	Functional Capacity (Peak VO <sub>2</sub> )	Cardiac Stress Biomarker (NT-proBNP)
Diabetes Stage A	~28 ml/kg/min	0-5 pg/ml (normal range)
DbCM Stage B	~25% decrease ≤20 ml/kg/min	~6-300 pg/ml
Stage C	>30% decrease 10-15 ml/kg/min	~300 - 5,000 pg/ml
Stage D	~24% of DbCM patients progress to overt heart failure or death within 1.5 years <sup>3</sup> 37% within 5 years <sup>4</sup>	> 5,000 pg/ml

LVH left ventricular hypertrophy, DbCM diabetic cardiomyopathy, HFpEF heart failure with preserved ejection fraction, HFrEF heart failure with reduced ejection fraction  
1. Kosmaia et al. J Am Coll Cardiol 2015;65:257-66; 2. Swank et al. Circ HF 2012; 3. Wang et al. JACC: Cardiovasc Imaging 2018; 4. From et al. JACC 2010

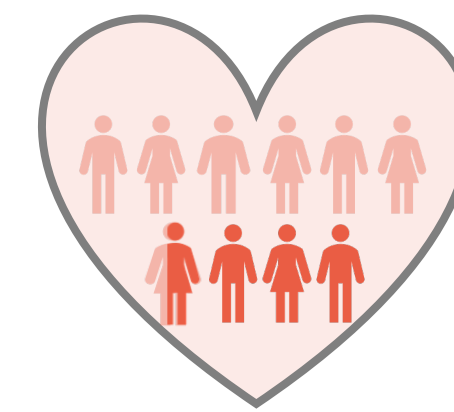
## High Prevalence of DbCM and Unmet Medical Need



Approximately, **17-24%** of patients with diabetes have DbCM in the absence of other forms of heart disease.<sup>1,2</sup>

~77 M patients worldwide have DbCM<sup>3</sup>

- ~ 8.0M in North America
- ~ 10.0M in Europe



- ~24% of DbCM patients progress to **overt heart failure or death** within 1.5 years<sup>4</sup>
- 37% within 5 years<sup>5</sup>

- Patients with diabetes are counseled on HF risk reduction:

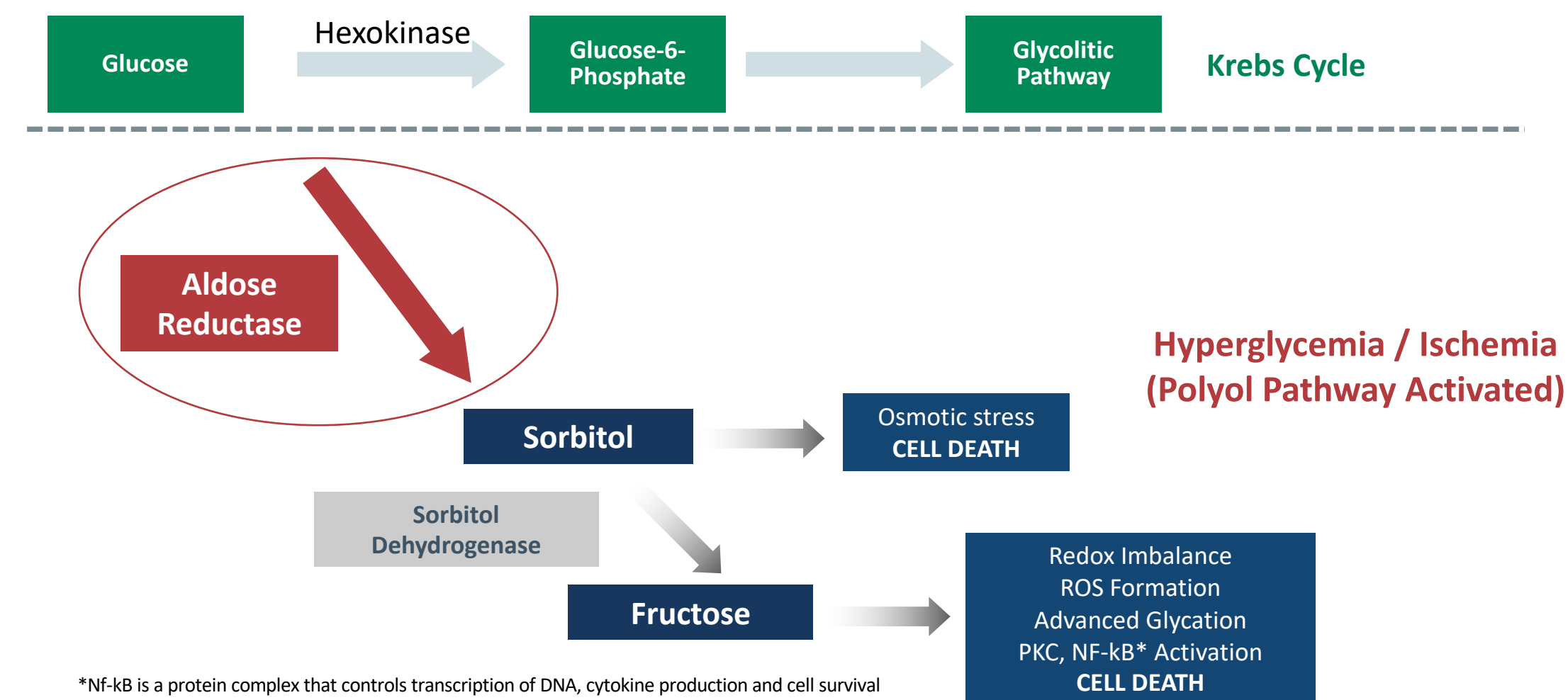
- Lifestyle modification
- Hypertension
- Dyslipidemia
- Hyperglycemia
- Albuminuria

### No Treatment for DbCM

- No therapies target the metabolic derangement responsible for DbCM and subsequent worsening to overt HF
- Heart Failure treatment is only initiated upon onset of clinical symptomatology (stage C heart failure)

1. Dandamudi et al. J Card Fail. 2014;20(5):304-309. 2. Pham et al. Intl J Endocrinology 3. International Diabetes Foundation, 2017.4. Wang et al. JACC: Cardiovasc Imaging 2018; 5. From et al. JACC 2010

## Pathogenesis of DbCM & Hyperactivation of Polyol Pathway<sup>1,2</sup>

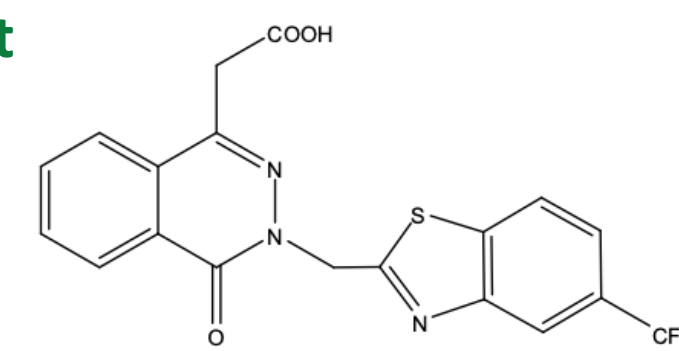


\*NF-kB is a protein complex that controls transcription of DNA, cytokine production and cell survival

1. Brownlee M. Diabetes Care. 2005;54(6):1615-1625. 2. Miki T, et al. Heart Fail Rev. 2013;18(2):149-166.

## AT-001 Pre-clinical and Clinical Profile Summary

### zopolrestat



Inhibition of Aldose Reductase

Clinical Efficacy

Competitive Inhibition of Aldehyde Reductase

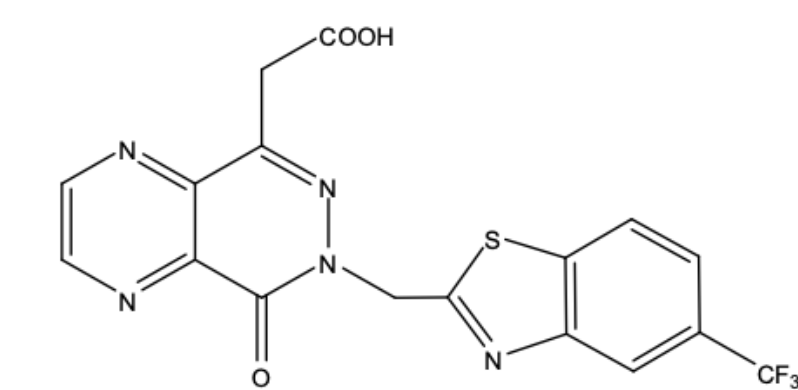
(Off-Target) Hepatotoxicity

- First generation Aldose Reductase Inhibitor (zopolrestat) demonstrated clinical efficacy in Diabetic Cardiomyopathy<sup>1</sup>
- Hepatotoxicity was observed in the development program (presumably due to off target competitive binding with Aldehyde Reductase in liver)
- Clinical development was discontinued

<sup>1</sup>Johnson, et al. Diabetes Care, 2004 pp 448-454

## AT-001: A Next Generation Highly Selective Aldose Reductase Inhibitor for Treatment of Diabetic Cardiomyopathy

### AT-001



- AT-001 was developed through rational drug design, using the geometric parameters of the active site of the Aldose Reductase enzyme determined via X-ray crystallography.
- Optimal target selectivity for Aldose Reductase and minimization of potential off-target activity with Aldehyde Reductase was achieved.
- Aldehyde Reductase plays an important role in detoxification mechanisms in the liver. Minimization of off-target activity is critical to ensure safety.

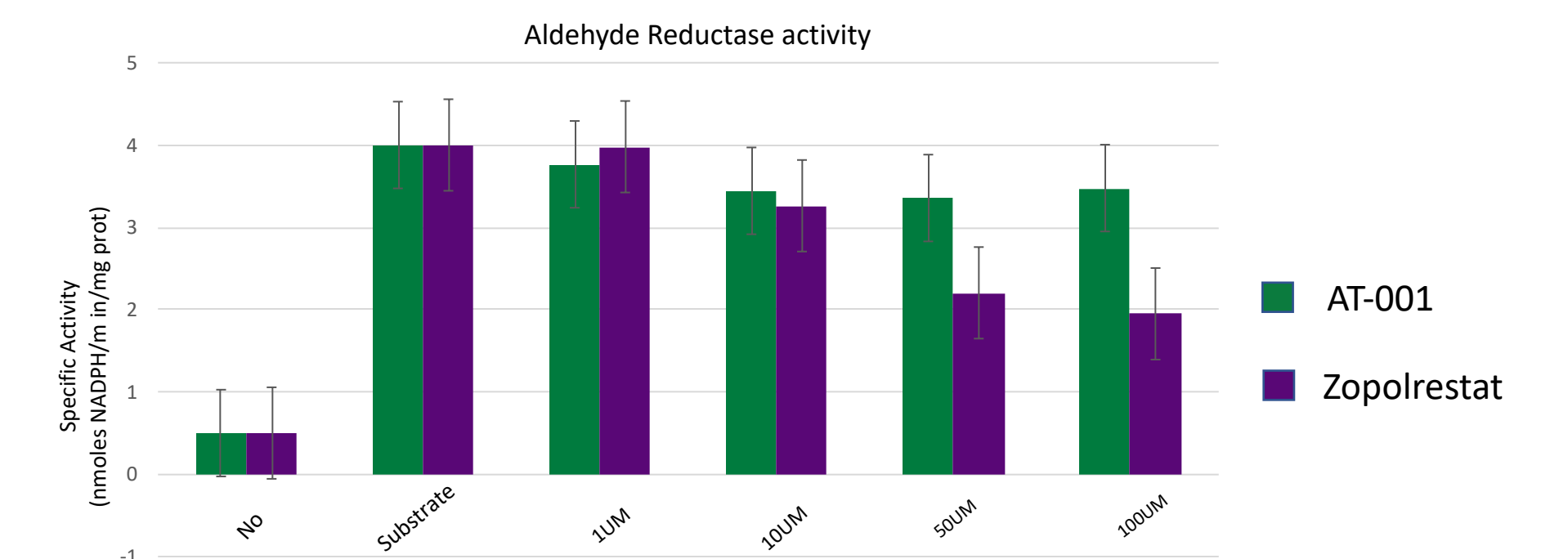
## AT-001 Increased Affinity for Aldose Reductase vs. Zopolrestat

Compound	Structure	IC <sub>50</sub>	MTD in animals	Tissue Penetration (in rats)			
				Syste mic/ Heart	Nerve	Retina	CNS
AT-001		30pM	>2,000mg/kg	✓	✓	✓	X
zopolrestat		10nM	100mg/kg	✓	✓	X	X

## No AT-001 Off-Target Binding

- Eurofins Panlabs Safety Screen Panel (consisting of 87 primary molecular targets including 13 enzyme and 74 binding assays) was used to evaluate potential off target binding activity of AT-001
- No off-target binding activity (defined as ≥50% inhibition or stimulation for biochemical assays) was observed

## Zopolrestat (But Not AT-001) Inhibits Aldehyde Reductase



## Conclusions

- AT-001 is logarithmically more potent than zopolrestat in inhibiting Aldose Reductase
- The unique structure and activity of AT-001 provide selectivity for Aldose Reductase and avoid off-target inhibition of Aldehyde Reductase
- The in vitro safety of this agent together with the positive safety data from the phase 1/2 program, support the ongoing pivotal study in DbCM