Supplementary Appendix

Supplement to: Desai AS, Webb DJ, Taube J, et al. Zilebesiran, an RNA interference therapeutic agent for hypertension. N Engl J Med 2023;389:228-38. DOI: 10.1056/NEJMoa2208391

This appendix has been provided by the authors to give readers additional information about the work.

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Participating Study Sites and List of Collaborators

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Co-investigators — Helen Wibberley, Mark Loughrey, Ben Hutter, Naomi How, James Mitchell, Andrew Stokes and Keith Berelowitz

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Co-investigators — Naimat Khan, Dave Singh

MAC Clinical Research, Blackpool, UK

Study Participants

Inclusion Criteria

- Male or female
- Age 18–65 years
- Mild-to-moderate hypertension (mean sitting systolic blood pressure [SBP] without anti-hypertensive medication >130 and ≤165 mmHg [Parts A and B]; or >135 and ≤165 mmHg [Part E]), by automated office blood pressure (AOBP) measurement
- Mean 24-hour SBP without anti-hypertensive medication by ambulatory monitoring ≥130 mmHg
- Untreated hypertension (not taking anti-hypertensive medication) or is on stable (no change in anti-hypertension medication or dose within 30 days prior to screening) monotherapy or dual therapy for hypertension with an angiotensin-converting enzyme inhibitor, angiotensin receptor blocker, calcium channel blocker, thiazide diuretic, or thiazide-like diuretic.
- Patients previously taking medication for hypertension must be without antihypertensives for ≥2 weeks prior to blood pressure measurement (4 weeks of washout is permitted for long-acting antihypertensive medications such as chlorthalidone and long-acting calcium channel blockers)
- 12-lead electrocardiogram (ECG) within normal limits or with no clinically significant abnormalities in the opinion of the investigator with a Fridericia-corrected QT interval (QTcF) of <450 ms in males or <470 ms in females
- Body mass index ≥18 and ≤35 kg/m^2 (Parts A and B); or ≥18 and ≤50 kg/m^2 (Part E)
- Written informed consent
- Females of child-bearing potential must be willing to use a highly effective method of contraception from 14 days before first dose, throughout study participation, and through safety follow-up until study completion

Exclusion Criteria

- Secondary hypertension
- Mean sitting diastolic blood pressure (DBP) >105 mmHg by AOBP measurement
- Orthostatic hypotension (fall of ≥20 mmHg SBP or ≥10 mmHg DBP within 1 minute of standing up from a supine position by AOBP)
- Clinically significant laboratory findings during screening
- Known active human immunodeficiency virus infection or evidence of current or chronic hepatitis C virus or hepatitis B virus infection
- Treatment within 30 days of randomization (Parts A and B) or 1 day of dosing (Part E) with a beta blocker, investigational agent, or medication that could impact patient safety or data integrity
- Treatment within 30 days of randomization (Parts A and B) or 1 day of dosing (Part E) with any herbal medicines, over-the-counter medications, or supplements known to affect blood pressure; chronic use of NSAIDs was not permitted
- Anticipated use of phosphodiesterase-5 inhibitors or organic nitrate preparations during course of study
- Diabetes mellitus
- History of:
    - Cardiovascular event
    - Multiple drug allergies or allergic reaction to an oligonucleotide or N-acetylgalactosamine-containing medication
    - Intolerance of subcutaneous (SC) injection(s)
- Any other medical condition or comorbidity that could impact patient compliance or data interpretation
- Clinically significant illness within 7 days before administration of study drug
- Weight loss of >10% in the last 6 months
- Estimated glomerular filtration rate of <60 mL/min/1.73 m^2

- Pregnancy (or planning a pregnancy) or lactation
- Unwillingness to comply with contraceptive requirements
- Unwillingness or inability to limit alcohol consumption to ≤2 units/day throughout the study
- Current alcohol or drug abuse, or within past 12 months
- Use of nicotine or tobacco-containing products within 6 months of study start
- Unwillingness to abstain from strenuous exercise for 48 hours before each blood collection
- Third-shift or night-shift workers

Study Design

Extended safety follow-up – Parts A, B, and E

After conclusion of the treatment period at 12 weeks for Parts A, B, and E, zilebesiran-treated patients continued to be monitored for safety until serum angiotensinogen levels reached either ≥50% (men) or ≥80% (women) of their individual baseline or until angiotensinogen levels were above the lower limit of normal. After the data cut-off on January 26, 2022, the protocol was updated to revise the duration of the safety follow-up period until angiotensinogen levels return to ≥50% of their individual baseline or until 12 months after their last dose of zilebesiran, whichever came earlier.

Part B: Single dose in a controlled salt intake cohort

The Part B cohort comprised 12 patients randomized 2:1 to receive a single SC dose of zilebesiran or placebo. The dose of zilebesiran did not exceed the highest dose that was found to have an acceptable safety profile in Part A.

Patients in Part B completed a 2-week dietary subprotocol that controlled sodium consumption to test for potential direct effects of salt-sensitive blood pressure. The low-salt diet (0.23 g/day) was administered on an inpatient setting on Days −21 to −15 and the high-salt diet (Days −14 to −8) on an outpatient setting. Each patient completed this dietary subprotocol once before study drug administration (Days −21 to −8) and once after study drug administration (Days 43 to 56). Patients were instructed to follow the study's general dietary recommendations (2.0 g sodium per day in the outpatient setting) from Day −7 to Day 42 and from Day 57 to study end.

Part C was planned as a multidose phase in hypertensive patients and was removed during a protocol amendment as the pharmacokinetic, pharmacodynamic, and safety data collected from Parts A and D were considered sufficient to inform future clinical studies of zilebesiran.

Part D: Multiple-dose phase in hypertensive patients who are obese

The Part D cohort comprised 12 patients with hypertension who have a body mass index of >35 kg/m^2 and ≤50 kg/m^2 in a single cohort. Patients were randomized 2:1 to receive zilebesiran or irbesartan in a double-blind, double dummy fashion as follows:

- Patients randomized to zilebesiran received two SC doses administered at Day 1 and Day 85 with once-daily oral doses of irbesartan-matching placebo administered in the morning
- Patients randomized to irbesartan received two SC doses of normal saline (0.9%) administered at Day 1 and Day 85 with once-daily oral doses of 150 mg irbesartan administered in the morning

This part of the study is ongoing; full details of the study methods and results will be published separately.

Part E: Single dose with irbesartan coadministration

Part E was conducted open label in up to 16 patients (or until at least eight patients received at least one dose of irbesartan) over 12 weeks. The dose of zilebesiran did not exceed the highest dose that was found to have an acceptable safety profile in Part A.

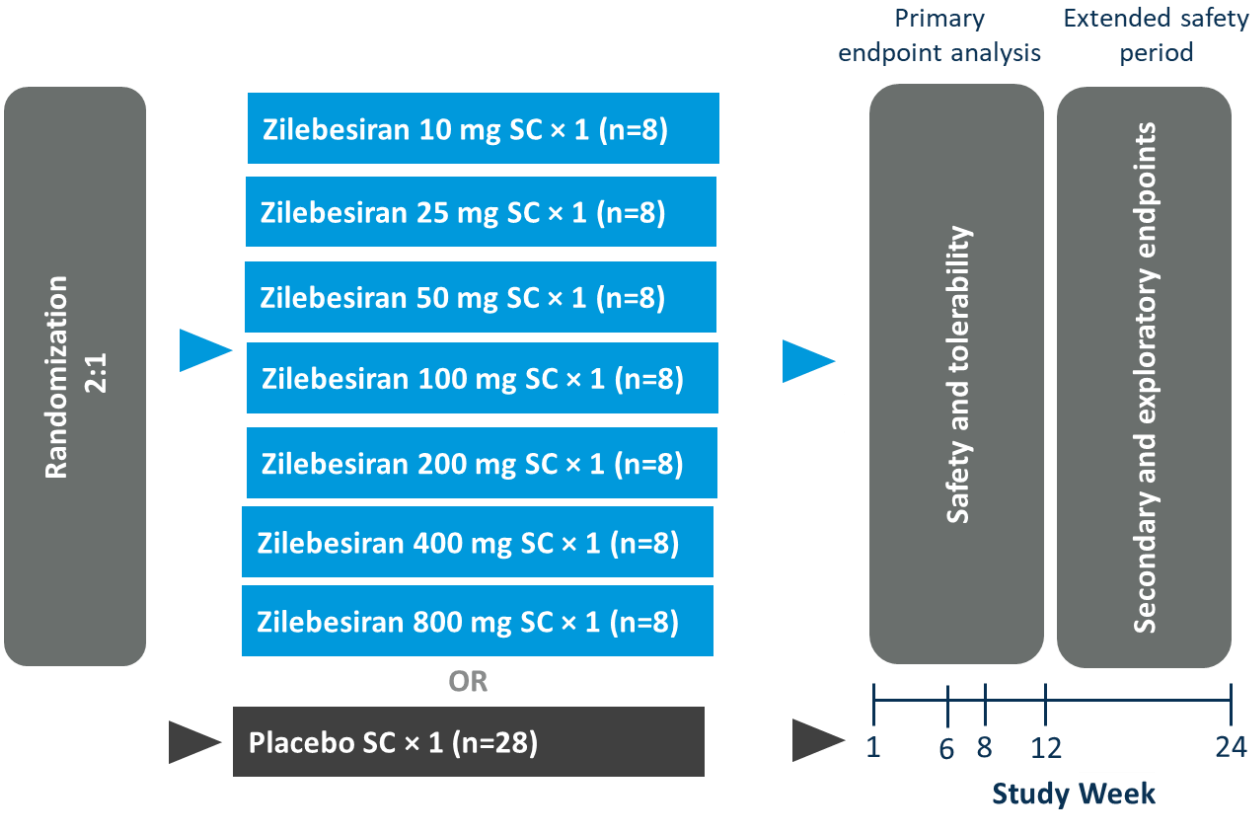
All patients received a single dose of zilebesiran on Day 1. If their 24-hour mean SBP on Day 41 remained at ≥120 mmHg, they received additional once-daily oral irbesartan 300 mg in the morning from Days 43 to 57.

Statistical Analysis

All patients who received any amount of zilebesiran or placebo were included in the safety analysis. All patients who received at least one dose of study drug and had at least one post-dose blood sample for the determination of serum angiotensinogen were included in the pharmacodynamics analysis. All patients who received at least one dose of study drug and had evaluable pharmacokinetic data from at least one post-dose blood or urine sample were included in pharmacokinetic assessments. The full analysis set, which was defined as all patients who received any amount of study drug, have baseline AOBP measurement, and have at least one post-baseline

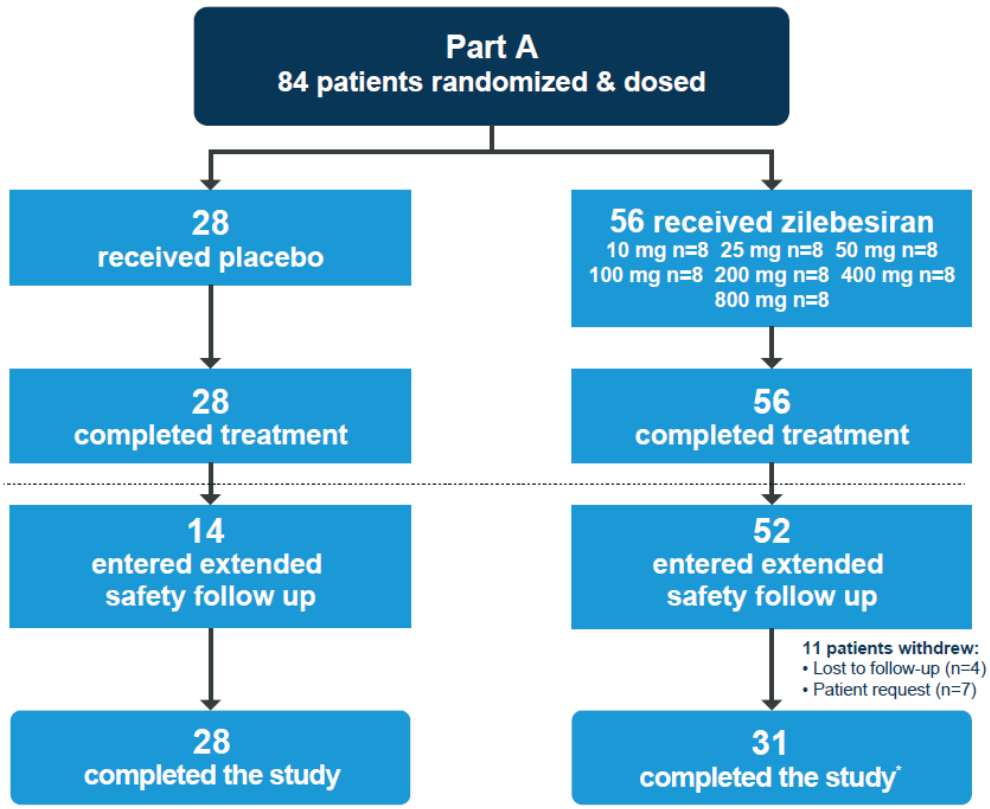
AOBP measurement, was used to assess exploratory efficacy by treatment group randomized (Parts A and B) or dosed (Part E).

Figure S1. Study Design in Patients with Hypertension (Part A).



Multicenter, randomized, double-blind, placebo-controlled, single ascending dose study. Each dose cohort included 12 patients (zilebesiran n=8, placebo n=4). After Week 8, if a patient developed clinically significantly elevated blood pressure, add-on treatment could be initiated at the discretion of the investigator and in line with current hypertension management guidelines. SC denotes subcutaneous

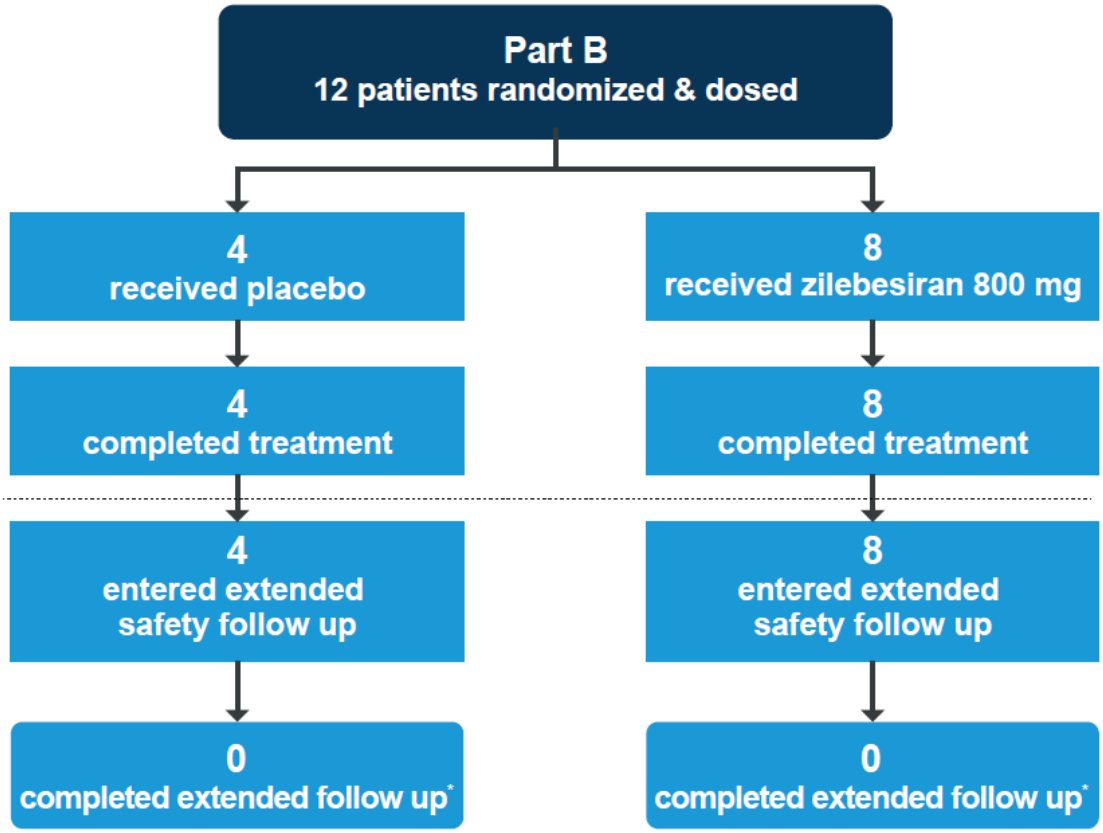
Figure S2. Patient Disposition.



A Part A

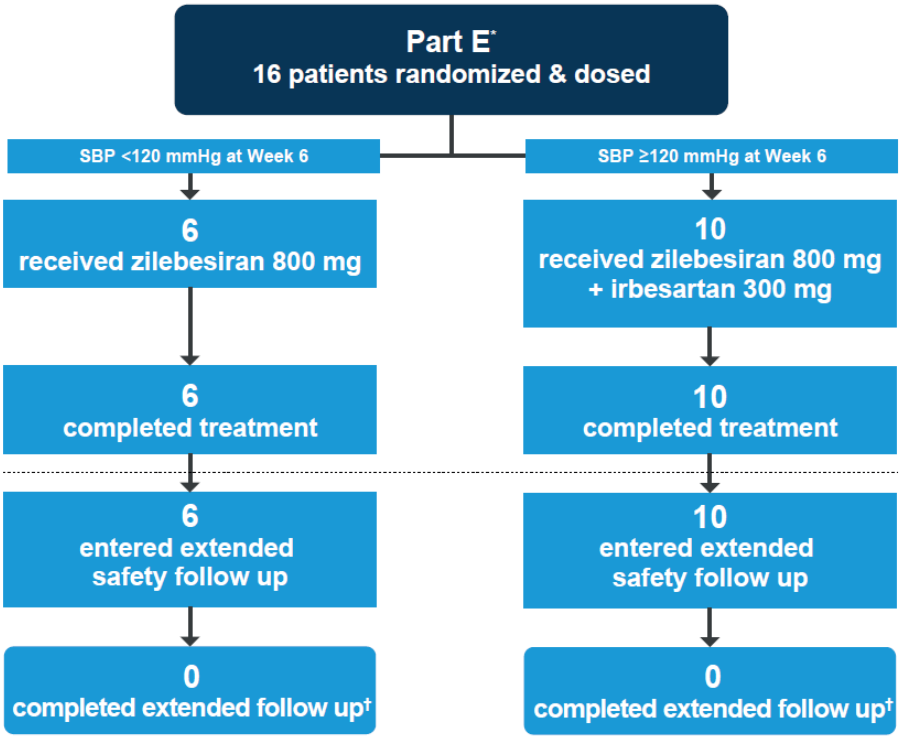
In Part A, eligible patients (12 per cohort) were randomly assigned 2:1 to receive a single subcutaneous dose of zilebesiran (10, 25, 50, 100, 200, 400, or 800 mg) or placebo. Add-on antihypertensive therapy was permitted at the discretion of the investigator at 8 weeks for uncontrolled hypertension. Study treatment period concluded at 12 weeks and after this, zilebesiran–treated patients continued to be monitored for safety until serum angiotensinogen levels had recovered to ≥50% (men) or ≥80% (women) of their individual baseline or until angiotensinogen levels are above the lower limit of normal. Data cut off was conducted on January 26, 2022 after the 12 week treatment period had completed, and all available safety data were assessed. *n=14 patients had not completed the study at data cut-off.

B Part B



In Part B, following sequential administration of low (0.23 g/day) and high (5.75 g/day) sodium diets (Days −21 to −8), patients were randomly assigned 2:1 to a single dose of zilebesiran 800 mg or placebo, and rechallenged with the same dietary protocol from Days 43–56. Study treatment period concluded at 12 weeks and after this, zilebesiran–treated patients continued to be monitored for safety until serum angiotensinogen levels had recovered to ≥50% (men) or ≥80% (women) of their individual baseline or until angiotensinogen levels are above the lower limit of normal. Data cut off was conducted on January 26, 2022 after the 12 week treatment period had completed, and all available safety data were assessed. *None of the patients had completed the extended safety follow-up period at data cut-off.

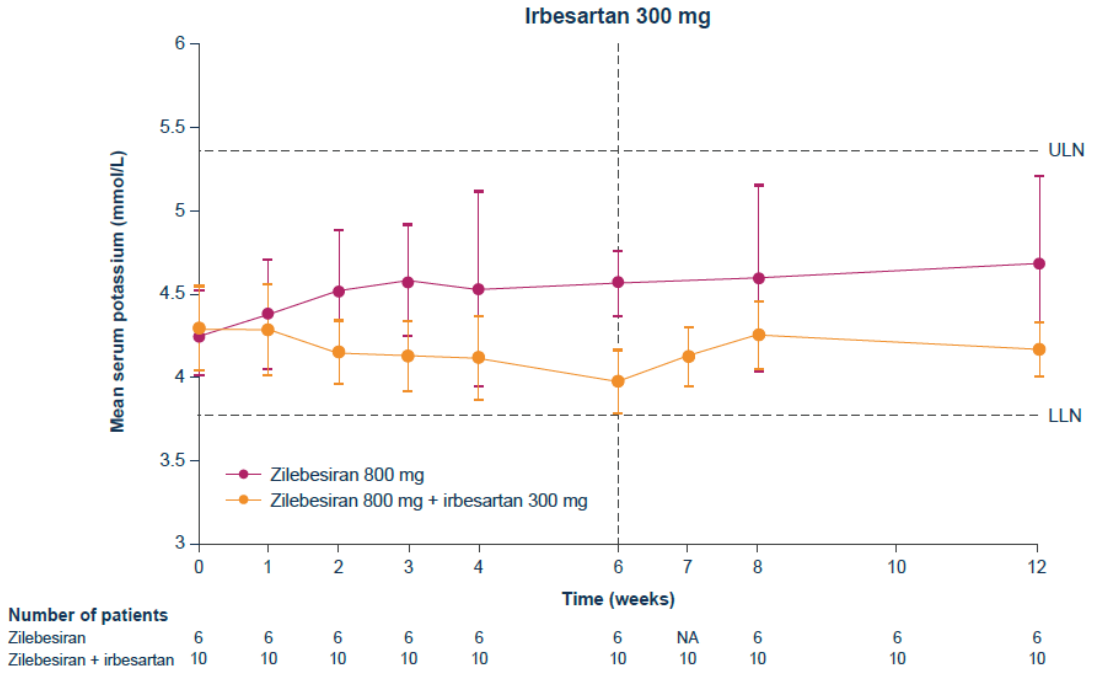
C Part E



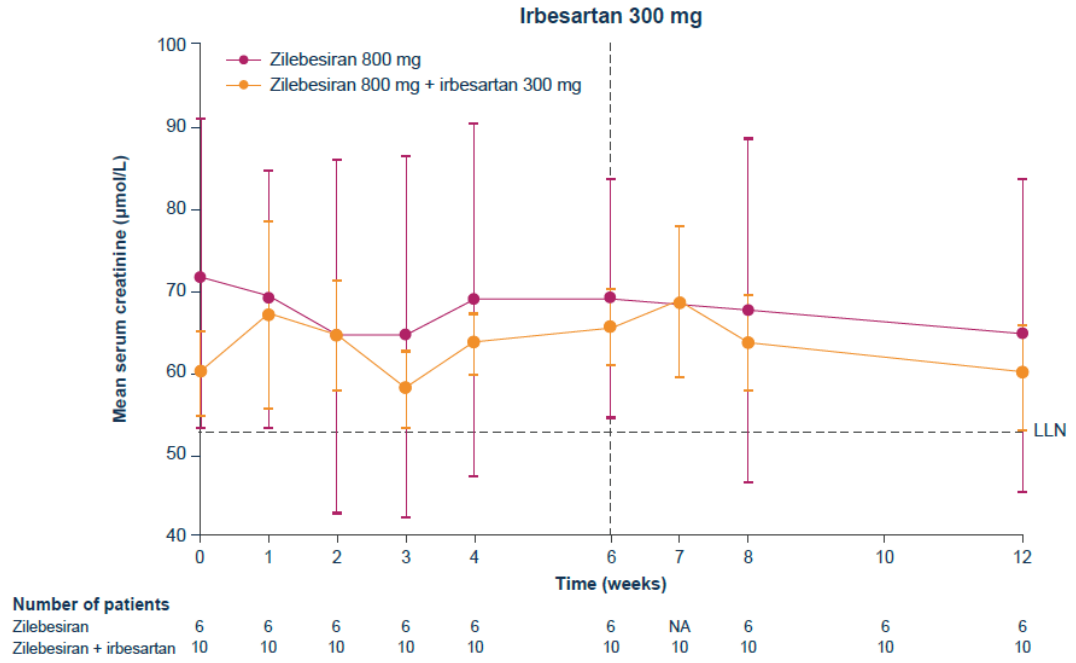
In Part E, all patients received a single dose of zilebesiran 800 mg. Patients with a systolic blood pressure ≥120 mmHg at Week 6 by 24-hour ambulatory blood pressure monitoring received additional treatment with irbesartan 300 mg once daily for 2 weeks. Study treatment period concluded at 12 weeks and after this, zilebesiran–treated patients continued to be monitored for safety until serum angiotensinogen levels had recovered to ≥50% (men) or ≥80% (women) of their individual baseline or until angiotensinogen levels are above the lower limit of normal. Data cut off was conducted on January 26, 2022 after the 12 week treatment period had completed, and all available safety data were assessed. *Part E included 5 patients who had participated in Part A and received placebo. †None of the patients had completed the extended safety follow-up period at data cut-off.

Figure S3. Serum Potassium and Creatinine Concentrations in Patients Receiving Zilebesiran with or without Add-On Irbesartan in Part E.

A Serum Potassium (mmol/L)

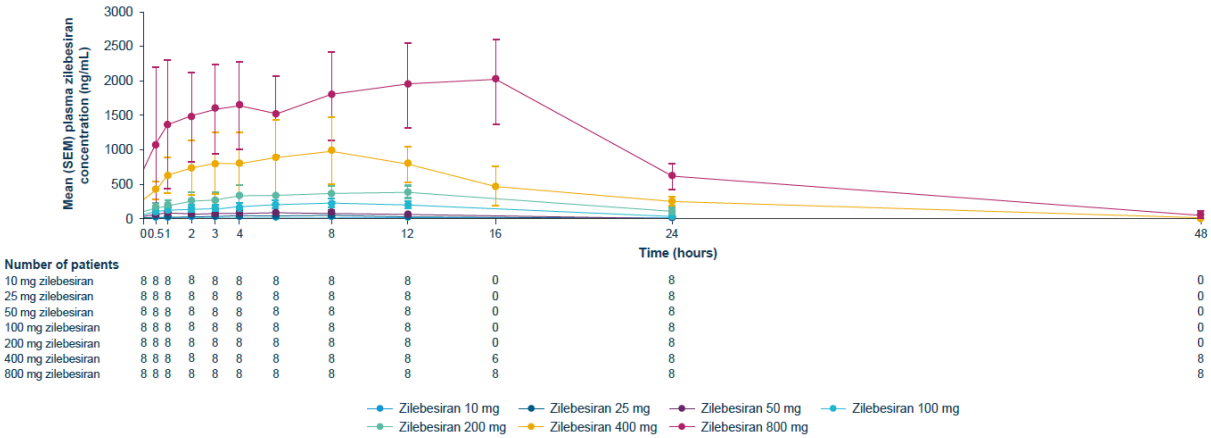


B Serum Creatinine (umol/L)



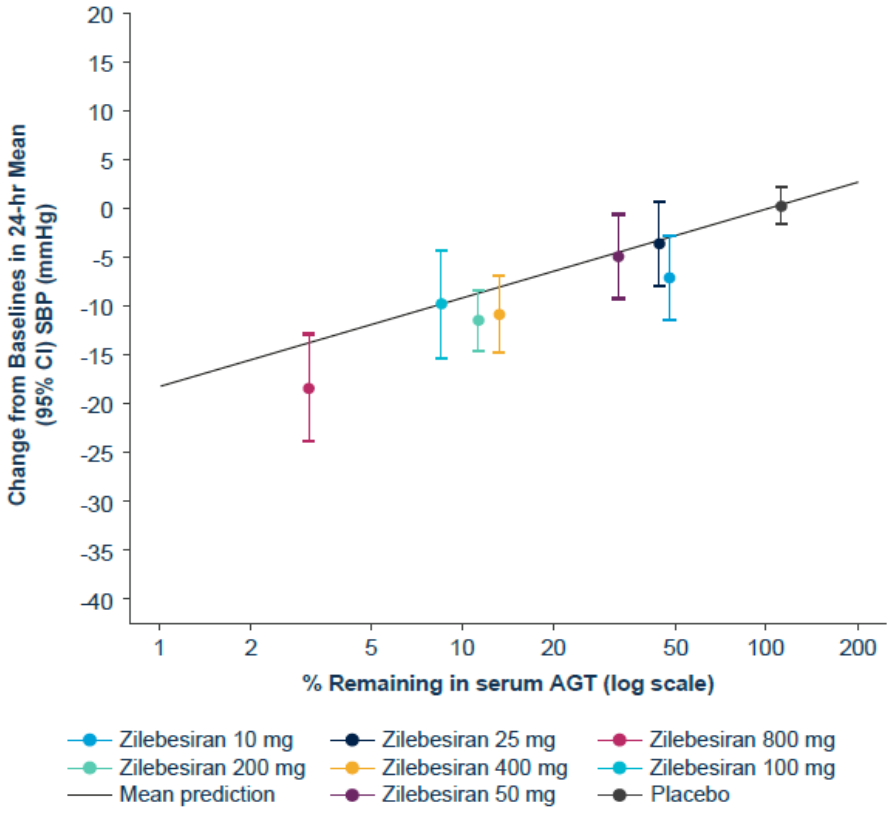
The graphs show the mean serum concentrations of potassium (Panel A) and creatinine (Panel B), over a period of 12 weeks, after administration of single doses of zilebesiran 800mg, with or without add-on irbesartan in Part E. In all panels, error bars are 95% confidence intervals (CI). Part E was a single-arm study with all patients receiving a single dose of zilebesiran 800 mg. Patients with a systolic blood pressure ≥120 mmHg at Week 6 by 24-hour ABPM received additional treatment with irbesartan 300 mg once daily for 2 weeks and are shown in the zilebesiran + irbesartan arm, with the dotted line indicating the timepoint irbesartan treatment commenced in these patients.

Figure S4. Mean Plasma Concentration of Zilebesiran in Patients with Hypertension after Receiving a Single Ascending Dose in Part A.



The graph depicts the mean zilebesiran plasma concentration over a 48-hour period after administration of single doses of zilebesiran in Part A, according to dose. Error bars are 95% confidence intervals (CI).

Figure S5. Model of the Relationship (log-normal) Between Serum AGT Reduction and 24-Hour Mean Systolic Blood Pressure Reduction (by ABPM) in Part A.



The figure depicts the relationship between AGT and blood pressure after administration of single doses of zilebesiran in Part A.

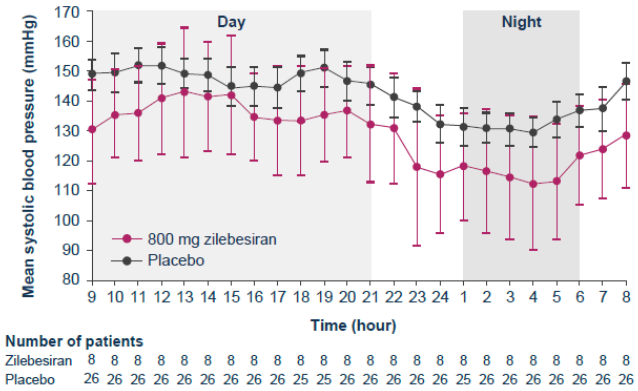
Solid black line represents model predicted line for the relationship (log-normal) between serum AGT reduction and 24-hour mean SBP reduction by ABPM. Closed circles and bars represent observed mean and 95% confidence intervals (CI) by dose group.

Figure contains all available SBP and AGT data from ALN-AGT01-001 Part A through 15 June 2021.

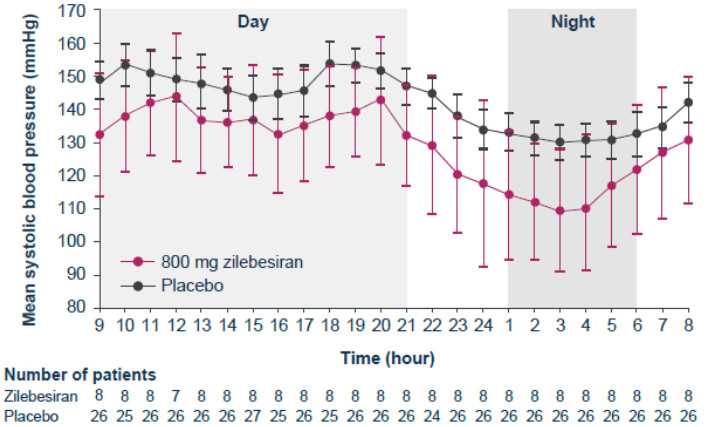
ABPM denotes ambulatory blood pressure monitoring, AGT angiotensinogen, CI, confidence intervals, and SBP systolic blood pressure.

Figure S6. Mean 24-hour Systolic Blood Pressure Following Single Dose of Zilebesiran 800 mg in Part A.

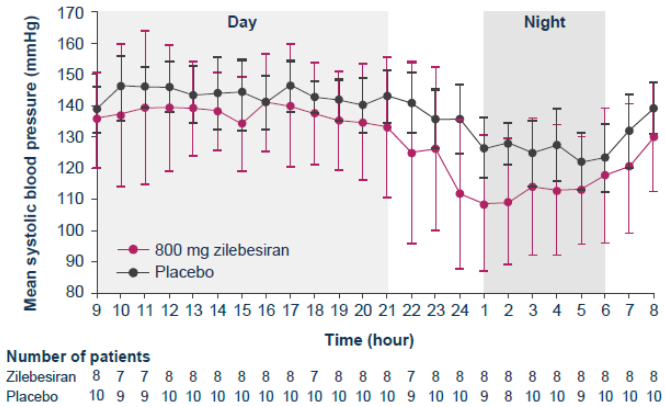
A Week 6: 24-hour SBP*



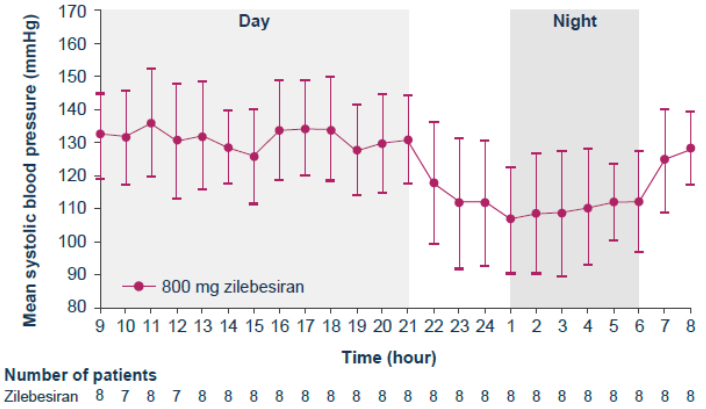
B Week 8: 24-hour SBP*†



C Week 12: 24-hour SBP*



D Week 24: 24-hour SBP*‡



The panels show the 24-hour systolic blood pressure, as an hourly adjusted mean, at Week 6 (Panel A), 8 (Panel B), 12 (Panel C), and 24 (Panel D) after administration of a single dose of zilebesiran 800mg or placebo in Part A. In all panels, the error bars represent 95% confidence intervals (CI). ABPM denotes ambulatory blood pressure monitoring, CI confidence interval, and SBP systolic blood pressure.

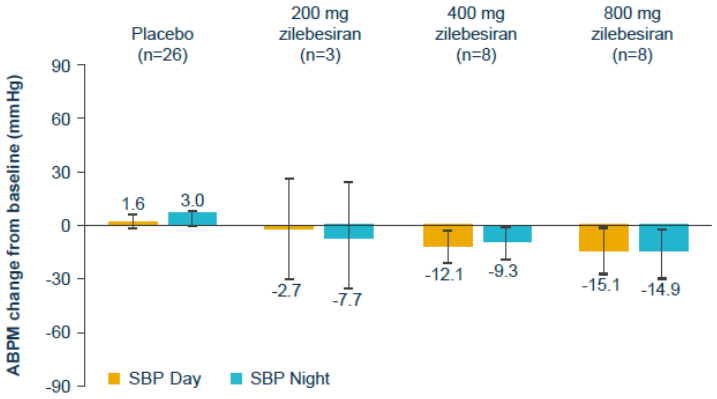
*Hourly adjusted mean; daytime [9 am to 9 pm], nighttime [1 am to 6 am].

†All patients who received zilebesiran 800mg did not require additional add-on antihypertensive medication up to Week 8.

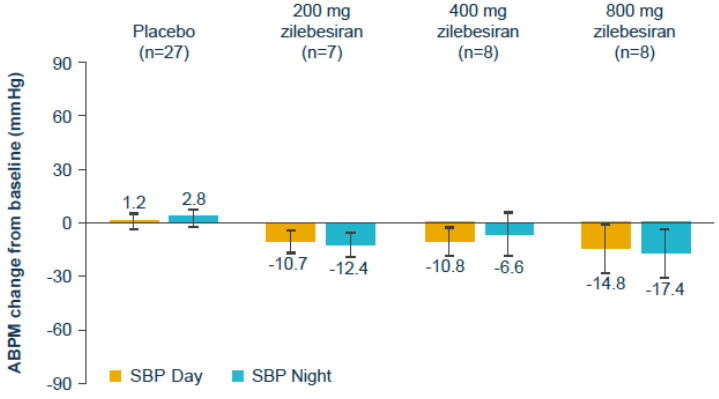
‡Patients receiving placebo were not required to enter the extended follow-up period (after Week 12).

Figure S7. Reductions in Daytime and Nighttime Systolic Blood Pressure Following Single Doses of Zilebesiran in Part A.

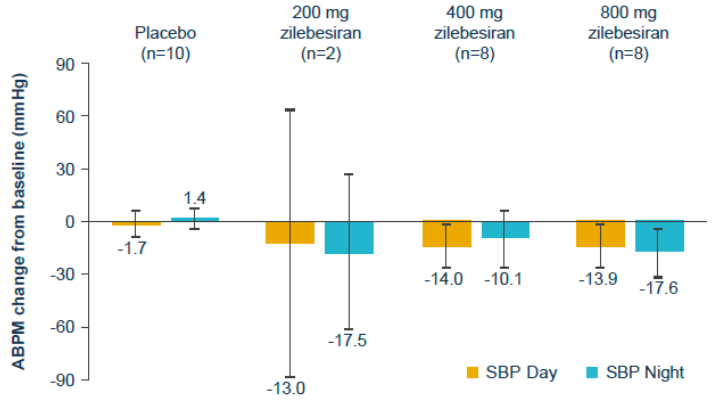
A Week 6*



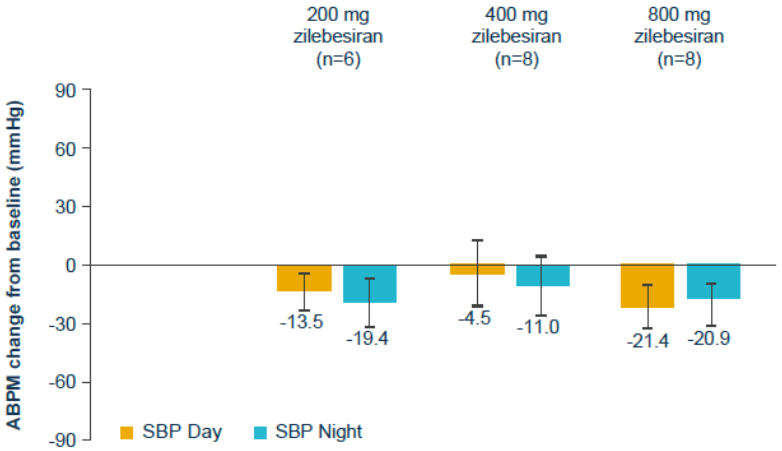
B Week 8*†



C Week 12



D Week 24‡§



The panels show change from baseline in the daytime and nighttime SBP at Week 6 (Panel A), 8 (Panel B), 12 (Panel C), and 24 (Panel D) after administration of a single dose of zilebesiran or placebo in Part A. Error bars are 95% confidence intervals (CI). ABPM denotes ambulatory blood pressure monitoring, CI confidence interval, and SBP systolic blood pressure.

Daytime included readings between 9 am to 9 pm, nighttime included readings between 1 am to 6 am. Mean baseline SBP/DBP: Placebo, 140.6/87.9 mmHg; zilebesiran 200 mg, 138.4/83.3 mmHg; 400 mg, 140.4/88.3 mmHg; 800 mg, 145.8/87.1 mmHg.

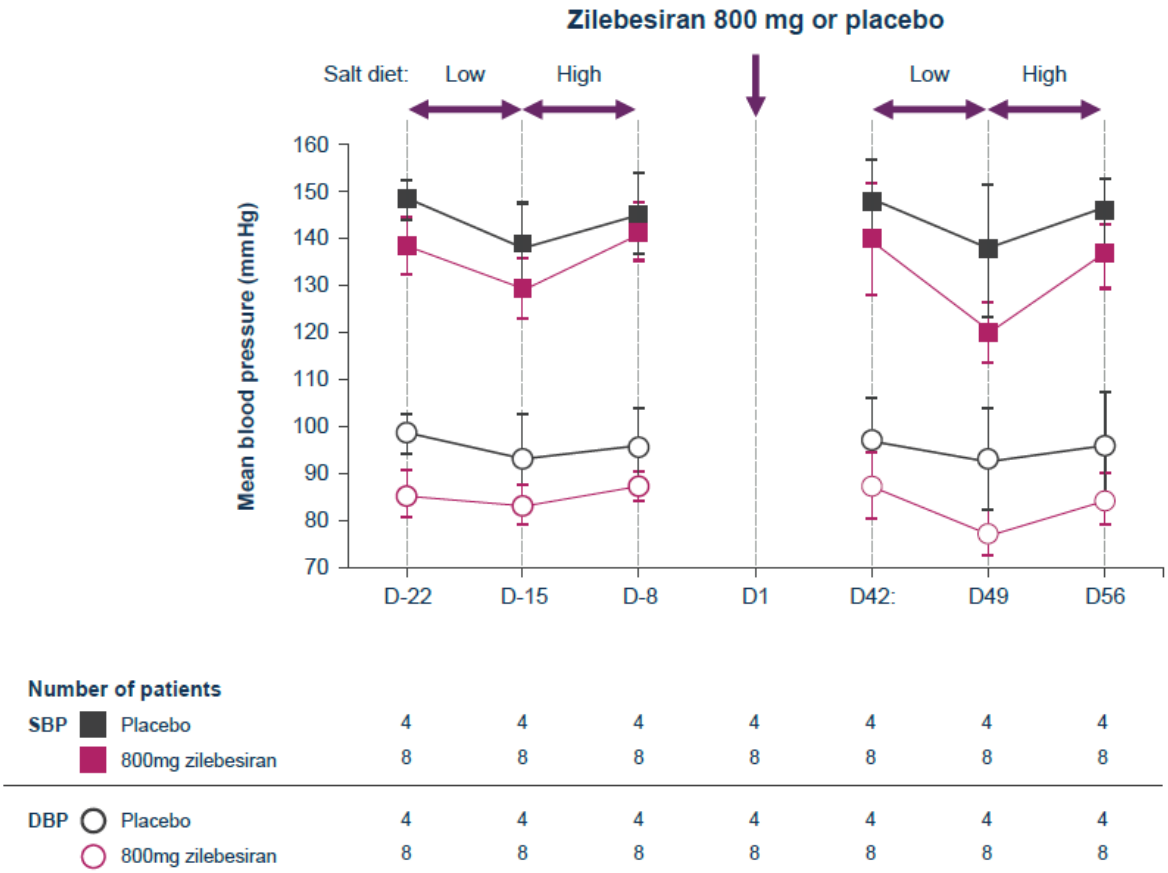
*During the study, add-on antihypertensive therapy was permitted at the discretion of the investigator for severe hypertension. One patient in the 200 mg dose group received add-on antihypertensive therapy before Week 8. After Week 8 add-on antihypertensive therapy was permitted at the discretion of the investigator for uncontrolled hypertension.

†In total, 26 patients were included in the SBP Night placebo arm and seven patients in the SBP Night 400 mg zilebesiran arm.

‡Five patients were included in the SBP Night 200 mg zilebesiran arm.

§Patients receiving placebo were not required to enter the extended follow-up period (after Week 12). Two patients in the 200 mg dose group, one patient in the 400 mg group, and two patients in the 800 mg group received add-on antihypertensive therapy.

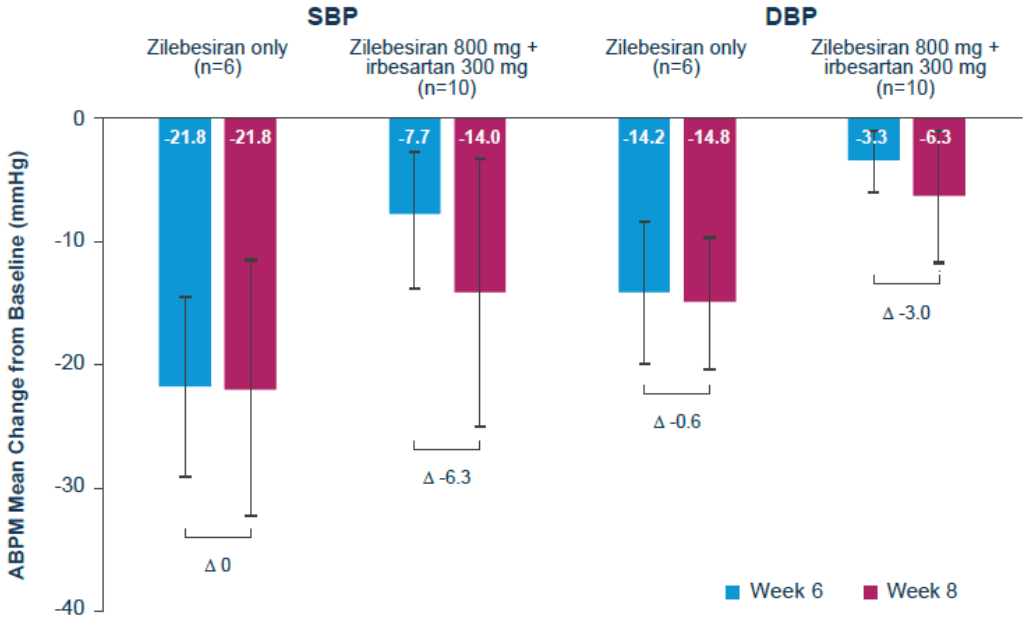
Figure S8. Change in Mean Blood Pressure from Day -22 to Day 56 in Patients with Hypertension Receiving Low and High Salt Diets in Part B.



In Part B, following sequential administration of low (0.23 g/day) and high (5.75 g/day) salt diets (Days −21 to −8), patients were randomly assigned 2:1 to a single dose of zilebesiran 800 mg or placebo, and rechallenged with the same dietary protocol from Days 43–56 (corresponding to the expected peak effect of zilebesiran). The panel shows the mean systolic and diastolic blood pressure during the dietary protocols before and after randomization. Error bars are 95% confidence intervals (CI).

D denotes day, DBP diastolic blood pressure, and SBP systolic blood pressure.

Figure S9. Mean Change from Baseline in Systolic and Diastolic Blood Pressure in Patients with Hypertension Receiving Zilebesiran Alone or Zilebesiran plus Irbesartan in Part E.*



The figure shows the mean changes from baseline in systolic and diastolic blood pressure at Week 6 and Week 8 after administration of single doses of zilebesiran, with or without irbesartan, in Part E. The error bars are 95% confidence intervals (CI).

*Part E was a single-arm study. At Week 6 all patients (n=16) had received a single dose of zilebesiran 800 mg only and had a mean (SEM) reduction in systolic blood pressure of 13.0 (2.6) mmHg. Patients with a systolic blood pressure ≥120 mmHg at Week 6 by 24-hour ABPM received additional treatment with irbesartan 300 mg once daily for 2 weeks (from Days 43 to 57) and are shown in the zilebesiran + irbesartan group. Week 6 data for the zilebesiran + irbesartan group are prior to treatment with irbesartan, whilst Week 8 data are following treatment with irbesartan.

ABPM denotes ambulatory blood pressure monitoring, DBP diastolic blood pressure, SBP systolic blood pressure, and SEM standard error of the mean.

Table S1: Representativeness of Study Participants

Category	Description
Disease under investigation	Hypertension
Special considerations related to:	
Sex and gender	• In the US and globally, the age-adjusted prevalence rates for hypertension are slightly higher in men than in women.[1-4] Rates of hypertension in premenopausal women tend to be lower than in men of similar age. As rates of hypertension in women tend to increase steeply after menopause the prevalence of hypertension tends to be higher in women than in men over the age of 65.[2,3,4]
Age	• The prevalence of hypertension in both men and women increases with age[2]
Race or ethnic group	• Hypertension disproportionately affects Black men and women in the United States[2,4,5] • Hypertension in Black patients tends to present earlier and is associated with higher risk for complications including stroke[3,6] and heart failure.
Geography	• The prevalence of hypertension varies globally by region, and is higher in low- and middle-income countries than in high-income countries.[1] • In men the lowest prevalence of hypertension is seen in South Asia and the highest prevalence in Eastern Europe and Central Asia. In women, the lowest prevalence is in high-income countries and the highest prevalence is in Sub-Saharan Africa. [1] • Rural residents have higher prevalence of hypertension than urban residents in high and middle-income countries, but the opposite pattern is seen in low-income countries. [4]
Other considerations	• Globally, it is estimated that 46% of adults who have hypertension are undiagnosed[7]
Overall representativeness of this trial	• All participants in this study were located in the United Kingdom • By design, the study excluded patients over the age of 65 to minimize the burden of comorbid medical illness in this phase 1 experience. Accordingly, the mean age of participants in the study was 53.5 years. • A higher proportion of men (62%) than women (38%) were enrolled, consistent with the higher population prevalence of hypertension in men than in women under the age of 65 years. • Despite the small study population, 25% of enrolled participants were identified as Black, which is comparable to other contemporary trials of hypertension

The data regarding hypertension epidemiology and demographics were synthesized from reports of US cardiovascular disease statistics published from the American Heart Association and National Health and Nutrition Survey and reports of global hypertension statistics from the World Health Organization. Data was supplemented from review of the key references cited in these documents as well as literature search using PubMed and concatenation of key terms of: 'hypertension', 'epidemiology', 'statistics', and 'United States' or 'global' or 'United Kingdom'. Specific references cited to support the epidemiologic observations noted are listed in the references section (at the end of this document).

Table S2. Summary of Adverse Events That Occurred during the Treatment Period among Patients Receiving Zilebesiran or Placebo per Dose in Part A.*

Outcome — no. (%)	Zilebesiran Dose							Total Part A	
	10 mg (n=8)	25 mg (n=8)	50 mg (n=8)	100 mg (n=8)	200 mg (n=8)	400 mg (n=8)	800 mg (n=8)	Placebo (N=28)	All Zilebesiran (N=56)
Adverse event	5 (62.5)	7 (87.5)	6 (75.0)	7 (87.5)	7 (87.5)	4 (50.0)	6 (75.0)	24 (85.7)	42 (75.0)
Any serious adverse event	0	0	0	0	1 (12.5)	0	0	1 (3.6)	1 (1.8)
Any severe adverse event	0	0	0	0	1 (12.5)	0	0	1 (3.6)	1 (1.8)
Any adverse event leading to withdrawal	0	0	0	0	0	0	0	0	0
Death	0	0	0	0	0	0	0	0	0
Adverse events occurring in ≥5% of patients									
Injection-site reaction	0	1 (12.5)	2 (25.0)	0	2 (25.0)	0	0	0	5 (8.9)
Upper respiratory tract infection	0	3 (37.5)	0	1 (12.5)	0	0	0	3 (10.7)	4 (7.1)
Myalgia	2 (25.0)	0	0	0	0	0	1 (12.5)	0	3 (5.4)
Dizziness	0	1 (12.5)	0	0	0	0	2 (25.0)	0	3 (5.4)
Headache	2 (25.0)	1 (12.5)	2 (25.0)	2 (25.0)	2 (25.0)	0	1 (12.5)	13 (46.6)	10 (17.9)
Oropharyngeal pain	0	1 (12.5)	2 (25.0)	0	0	0	0	0	3 (5.4)

*Adverse events occurring in ≥5% patients are shown based on the adverse events ≥5% in the all zilebesiran group.

Serious adverse events were defined as adverse events that resulted in death, were life-threatening, required inpatient hospitalization, or prolongation of existing hospitalization, resulted in persistent or significant disability or incapacity, were a congenital anomaly or birth defect, or were important medical events as determined by the investigators. All adverse events (including serious adverse events) were graded for severity. Severe events were adverse events for which more than minimal, local, or noninvasive intervention was indicated; had a severe effect on limiting self-care activities of daily living; or had potential for life-threatening consequences or death. Serious adverse events included a severe event of optic ischemic neuropathy (placebo) and a severe event of prostate cancer (zilebesiran 200 mg) based on biopsy during screening. There were no deaths or unplanned hospitalizations, and no patients required intervention for symptomatic hypotension, hyperkalemia, or worsening renal function.

Table S3. Changes in Kidney Function and Electrolytes from Baseline to Week 12 in Parts A, B, and E

Parameter	Part A		Part B		Part E	
	Placebo	Zilebesiran All Doses	Placebo	Zilebesiran 800 mg	Zilebesiran 800 mg	Zilebesiran 800 mg + Irbesartan 300 mg
Serum creatinine concentration (µmol/L)						
Baseline						
N	28	56	4	8	6	10
Mean (95% CI)	71.1 (66.6, 75.7)	70.7 (66.8, 74.6)	62.0*	69.8 (60.6, 78.9)	72.8 (53.9, 91.8)	60.2 (55.1, 65.3)
Week 12						
N	28	53	2	2	6	10
Mean (95% CI)	67.1 (62.4, 71.8)	69.8 (66.1, 73.5)	62.0 (−52.4, 176.4)	57.5 (0.3, 114.7)	64.8 (45.6, 84.1)	60.2 (53.6, 66.8)
Change from baseline at Week 12	n=28	n=53	n=2	n=2	n=6	n=10
Mean (95% CI)	−4.0 (−6.9, −1.2)	−0.2 (−2.2, 1.9)	0.0 (−114.4, 114.4)	−4.5 (−61.7, 52.7)	−8.0 (−12.5, −3.5)	0.0 (−4.3, 4.3)
Estimated glomerular filtration rate (mL/min/1.73 m^2)						
Baseline						
N	28	56	4	8	5	10
Mean (95% CI)	95.2 (90.4, 100.0)	100.3 (95.1, 105.5)	116.5 (112.3, 120.7)	100.6 (87.4, 113.9)	98.4 (63.0, 133.8)	108.0 (95.4, 120.6)
Week 12						
N	28	53	N/A†	N/A†	6	10
Mean (95% CI)	102.6 (96.3, 108.9)	100.8 (96.1, 105.5)			114.3 (79.1, 149.6)	109.8 (90.7, 129.0)
Change from baseline at Week 12	n=28	n=53	N/A†	N/A†	n=5	n=10
Mean (95% CI)	7.4 (1.5, 13.3)	−0.2 (−4.1, 3.7)			13.0 (−2.2, 28.2)	1.8 (−7.4, 11.0)

Serum potassium (mmol/L)						
Baseline						
N	28	56	4	8	6	10
Mean (95% CI)	4.4 (4.3, 4.5)	4.4 (4.3, 4.5)	4.4 (3.9, 4.8)	4.3 (4.0, 4.6)	4.3 (4.0, 4.5)	4.3 (4.0, 4.6)
Week 12						
N	28	54	2	2	6	10
Mean (95% CI)	4.3 (4.1, 4.4)	4.4 (4.3, 4.5)	4.7 (−1.1, 10.4)	4.1 (−1.0, 9.2)	4.7 (4.2, 5.2)	4.2 (4.0, 4.3)
Change from baseline at Week 12	n=28	n=54	n=2	n=2	n=6	n=10
Mean (95% CI)	−0.1 (−0.3, 0.1)	−0.0 (−0.1, 0.1)	0.2 (−1.8, 2.1)	0.1 (−2.4, 2.6)	0.4 (0.1, 0.8)	−0.1 (−0.4, 0.1)
Serum sodium (mmol/L)						
Baseline						
N	28	56	4	8	6	10
Mean (95% CI)	139.4 (138.7, 140.1)	139.9 (139.4, 140.4)	140.3 (135.7, 144.8)	139.6 (138.2, 141.03)	138.2 (134.9, 141.4)	138.5 (136.7, 140.3)
Week 12						
N	28	54	2	2	6	10
Mean (95% CI)	139.2 (138.3, 140.0)	139.4 (138.9, 140.0)	140.5 (134.1, 146.9)	141.5 (135.1, 147.9)	137.0 (134.7, 139.3)	139.4 (137.8, 141.0)
Change from baseline at Week 12	n=28	n=53	n=2	n=2	n=6	n=10
Mean (95% CI)	−0.2 (−0.9, 0.4)	−0.5 (−1.1, 0.1)	−1.5 (−7.9, 4.9)	2.5 (−3.9, 8.9)	−1.2 (−4.5, 2.2)	0.9 (−1.4, 3.2)

*No 95% CI, as all patient values were identical.

†Week 8 estimated glomerular filtration rate (mL/min/1.73 m^2) data are as follows:

Part B — Placebo (N=4): mean (95% CI) 118.5 (88.2, 148.8); change from baseline at Week 8 (n=4): mean (95% CI) 2.0 (−24.2, 28.2)

Zilebesiran 800 mg (N=6): mean (95% CI) 104.5 (91.7, 117.3); change from baseline at Week 8 (n=6): mean (95% CI) 2.3 (−9.9, 14.6).

CI denotes confidence interval.

Table S4. Changes in Bodyweight Following Single Doses of Zilebesiran in Part A.

Bodyweight (kg)	Placebo	Zilebesiran Dose						
		10 mg	25 mg	50 mg	100 mg	200 mg	400 mg	800 mg
Baseline	n=28	n=8	n=8	n=8	n=8	n=8	n=8	n=8
Mean (95% CI)	86.62 (82.12. 91.12)	82.95 (70.76, 95.14)	77.01 (70.06, 83.96)	87.90 (74.64, 101.16)	80.61 (71.68, 89.54)	86.75 (77.34, 96.16)	87.98 (73.73, 102.22)	84.39 (69.01, 99.76)
Week 12	n=28	n=7	n=8	n=7	n=7	n=8	n=8	n=8
Mean (95% CI)	87.23 (82.51, 91.96)	84.50 (71.09, 97.91)	75.93 (69.47, 82.39)	86.40 (72.48, 100.32)	81.71 (72.42, 91.01)	88.83 (79.28, 98.37)	89.38 (75.29, 103.46)	86.69 (70.09, 103.28)
Change from baseline at Week 12	n=28	n=7	n=8	n=7	n=7	n=8	n=8	n=8
Mean (95% CI)	0.61 (−0.18, 1.40)	1.55 (−0.24, 3.34)	0.77 (−1.14, 2.68)	1.23 (−0.24, 2.70)	1.97 (0.84, 3.10)	2.08 (0.68, 3.47)	1.40 (−0.71, 3.51)	2.30 (0.07, 4.53)

CI denotes confidence interval.

Table S5. Changes in RAAS biomarkers from Baseline to Week 12 in Part A

Parameter		Zilebesiran Dose						
	Placebo	10 mg	25 mg	50 mg	100 mg	200 mg	400 mg	800 mg
	(n=28)	(n=8)	(n=8)	(n=8)	(n=8)	(n=8)	(n=8)	(n=8)
Aldosterone concentration (nmol/L)								
Baseline	n=28	n=8	n=8	n=8	n=8	n=8	n=8	n=8
Mean (95% CI)	0.370 (0.312, 0.427)	0.265 (0.193, 0.337)	0.352 (0.173, 0.531)	0.344 (0.245, 0.443)	0.319 (0.219, 0.419)	0.417 (0.327, 0.508)	0.340 (0.287, 0.393)	0.371 (0.274, 0.469)
Week 12	n=25	n=8	n=8	n=7	n=7	n=8	n=8	n=3
Mean (95% CI)	0.457 (0.367, 0.548)	0.366 (0.291, 0.441)	0.371 (0.215, 0.527)	0.477 (0.170, 0.785)	0.259 (0.176, 0.342)	0.317 (0.213, 0.422)	0.270 (0.195, 0.344)	0.197 (0.051, 0.342)
Change from baseline to Week 12	n=25	n=8	n=8	n=7	n=7	n=8	n=8	n=3
Mean (95% CI)	0.092 (0.002, 0.181)	0.101 (0.064, 0.138)	0.019 (-0.154, 0.191)	0.142 (-0.084, 0.368)	−0.037 (-0.135, 0.062)	−0.100 (−0.200, −0.001)	−0.071 (−0.142, 0.001)	−0.094 (−0.336, 0.149)
Angiotensin I concentration (pmol/L)								
Baseline	n=28	n=8	n=8	n=8	n=8	n=8	n=8	n=8
Mean (95% CI)	4.729 (3.255, 6.202)	2.756 (1.583, 3.930)	2.975 (1.671, 4.279)	4.781 (1.975, 7.588)	2.419 (1.585, 3.253)	6.950 (3.716, 10.184)	5.731 (2.996, 8.467)	5.631 (2.839, 8.423)
Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=6

Mean (95% CI)	7.193 (4.007, 10.378)	3.138 (1.601, 4.674)	4.150 (−0.366, 8.666)	6.286 (0.002, 12.569)	3.914 (0.388, 7.441)	2.525 (1.682, 3.368)	2.250 (1.884, 2.616)	2.617 (2.084 3.149)
Change from baseline to Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=6
Mean (95% CI)	2.472 (0.106, 4.838)	0.381 (−1.384, 2.146)	1.175 (−3.512, 5.862)	1.157 (−3.593, 5.908)	1.436 (−2.467, 5.338)	−4.425 (−7.086, −1.764)	−3.481 (−5.947, −1.015)	−3.342 (−6.764, 0.080)
Angiotensin II concentration (pmol/L)								
Baseline	n=28	n=8	n=8	n=8	n=8	n=8	n=8	n=8
Mean (95% CI)	3.230 (2.178, 4.283)	2.494 (1.806, 3.181)	2.500 (1.847, 3.153)	3.156 (1.882, 4.430)	2.088 (1.881, 2.294)	2.825 (2.053, 3.597)	3.094 (2.077, 4.111)	4.794 (2.195, 7.393)
Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=6
Mean (95% CI)	7.152 (2.241, 12.063)	2.775 (2.051, 3.499)	3.188 (0.380, 5.995)	2.857 (1.465, 4.249)	2.300 (1.566, 3.034)	2.263 (1.642, 2.883)	2.363 (1.929, 2.796)	2.033 (1.948, 2.119)
Change from baseline to Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=6
Mean (95% CI)	3.896 (−0.670, 8.462)	0.281 (−0.358, 0.921)	0.688 (−2.179, 3.554)	−0.464 (−2.182, 1.253)	0.200 (−0.612, 1.012)	−0.563 (−1.359, 0.234)	−0.731 (−1.397, −0.065)	−3.075 (−6.703, 0.553)
Plasma renin activity (ng/mL/h)								
Baseline	n=28	n=8	n=8	n=8	n=8	n=8	n=8	n=8
Mean (95% CI)	0.308 (0.176, 0.439)	0.266 (0.073, 0.459)	0.166 (−0.030, 0.362)	0.330 (0.055, 0.605)	0.079 (0.013, 0.146)	0.273 (0.021, 0.525)	0.414 (0.209, 0.620)	0.428 (0.033, 0.822)

Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=8
Mean (95% CI)	0.489 (0.195, 0.782)	0.261 (0.051, 0.472)	0.286 (−0.234, 0.806)	0.287 (−0.073, 0.647)	0.084 (−0.122, 0.291)	0.058 (−0.033, 0.148)	0.124 (−0.005, 0.252)	0.020 (−0.024, 0.064)
Change from baseline to Week 12	n=27	n=8	n=8	n=7	n=7	n=8	n=8	n=8
Mean (95% CI)	0.182 (−0.044, 0.409)	−0.005 (−0.179, 0.169)	0.120 (−0.381, 0.621)	−0.071 (−0.453, 0.312)	0.009 (−0.184, 0.201)	−0.216 (−0.473, 0.042)	−0.291 (−0.464, −0.118)	−0.408 (−0.765, −0.050)

RAAS denotes renin–angiotensin–aldosterone system, CI denotes confidence interval.

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