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A novel model of cardiovascular–kidney–metabolic syndrome combining unilateral nephrectomy and high-salt–sugar–fat diet in mice

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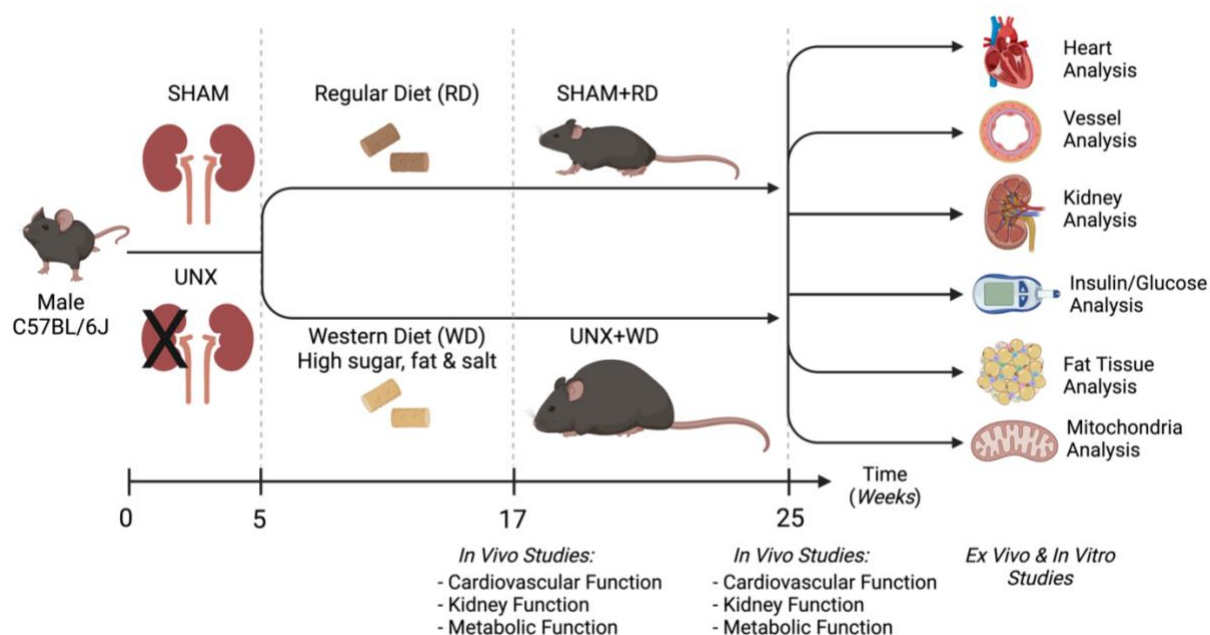
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SUPPLEMENTARY INFORMATION



Supplementary Figure 1: Schematic of experimental design for a model of cardiovascular-kidney-metabolic (CKM) syndrome in mice. Male C57BL/6J mice (4 weeks old) underwent unilateral nephrectomy (UNX) and were subsequently fed a customized Western diet (WD) high in sugar, fat, and salt for 12 or 20 weeks. Sham-operated mice, fed regular rodent diet for the same durations, served as healthy controls (SHAM+RD). *In vivo* studies assessed cardiovascular function (blood pressure and echocardiography), kidney function (glomerular filtration rate), and metabolic functions (body composition, weight gain, glucose metabolism, fat distribution). These *in vivo* assessments were followed by *ex vivo* functional and *in vitro* mechanistic studies, which included isolated vessel reactivity, mitochondrial respiration, histopathology, and biochemical analysis of various tissues. Illustration created with BioRender: Carlstrom, M. (2024) BioRender.com/s35e756.

SUPPLEMENTARY INFORMATION

Supplementary Table 1. Composition of special Western diet.

Class description	Ingredients	Grams (g)
Protein	Casein, Lactic, 30 Mesh	200.00
Protein	Methionine, DL	3.00
Carbohydrate	Sucrose, Fine Granulated	500.00
Carbohydrate	Starch, Corn	150.00
Fiber	Solka Floc, FCC200	50.00
Fat	Corn Oil	50.00
Mineral	Sodium Chloride	42.00
Mineral	S10001	35.00
Vitamin	V10001	10.00
Vitamin	Choline Bitartrate	2.00
Dye	Dye, Green FD&C #40	0.05
Total		1042.05 g

Detailed formulation of the customized Western diet rich in sugar, fat, and salt (WD) produced by Research Diets, Inc. (New Brunswick, NJ, 08901 USA), Product code D06111702-1.5Vii. This product is a modified version of Research Diet's traditional WD (D12079B-1.5Vii).

SUPPLEMENTARY INFORMATION

Supplementary Table 2. Description of sample numbers.

	SHAM+RD	UNX+WD 12 weeks	UNX+WD 20 weeks
Fig 1A	8	7	8
Fig 1B	12	12	8
Fig 1C	12	12	6
Fig 1D	12	12	8
Fig 1E	12	4	8
Fig 1F	12	4	8
Fig 3A	12	12	8
Fig 3B	12	8	8
Fig 3C	12	8	8
Fig 3D	8	8	8
Fig 3E	8	8	8
Fig 3F	8	8	8
Fig 3G	11	7	5
Fig 3H	10	8	5
Fig 3I	12	12	8
Fig 4A	6	7	8
Fig 4B	6	7	8
Fig 4C	6	7	8
Fig 4D	12	12	8
Fig 4E	9	10	6
Fig 4F	6	7	8
Fig 5A	8	8	8
Fig 5B	12	12	8
Fig 5C	40*	41*	39*
Fig 6A	12	12	8
Fig 6B	12	12	8
Fig 6C	11	11	7
Fig 6D	11	8	6

Description of sample numbers by methods used in figures 1, 3, 4, 5 and 6. *Number of photomicrographs analyzed - average of 4 photomicrographs per tissue sample. Abbreviations: RD, regular diet; UNX, unilateral nephrectomy; WD, Western diet.