

Special Section on Non-Coding RNAs in Clinical Practice: From Biomarkers to Therapeutic Tools

Empagliflozin Improves the MicroRNA Signature of Endothelial Dysfunction in Patients with Heart Failure with Preserved Ejection Fraction and Diabetes

Pasquale Mone, Angela Lombardi, Urna Kansakar, Fahimeh Varzideh, Stanislovas S. Jankauskas, Antonella Pansini, Stefania Marzocco, Stefano De Gennaro, Michele Famiglietti, Gaetano Macina, Salvatore Frullone, and Gaetano Santulli

Department of Medicine, Fleischer Institute for Diabetes and Metabolism (FIDAM), Einstein Institute for Aging Research, Einstein-Mount Sinai Diabetes Research Center (ES-DRC), Albert Einstein College of Medicine, New York City, New York (P.M., A.L., U.K., F.V., S.S.J., G.S.); Azienda Sanitaria Locale (ASL) Avellino, Avellino, Italy (P.M., A.P., S.D.G., M.F., G.M., S.F.); University of Salerno, Fisciano, Italy (S.M.); International Translational Research and Medical Education Consortium (ITME) and Department of Advanced Biomedical Sciences, “Federico II” University, Naples, Italy (G.S.); and Department of Molecular Pharmacology, Wilf Family Cardiovascular Research Institute, Institute for Neuroimmunology and Inflammation, Albert Einstein College of Medicine, New York City, New York (U.K., F.V., S.S.J., G.S.)

Received September 8, 2021; accepted June 6, 2022

ABSTRACT

Endothelial dysfunction represents a key mechanism underlying heart failure with preserved ejection fraction (HFpEF), diabetes mellitus (DM), and frailty. However, reliable biomarkers to monitor endothelial dysfunction in these patients are lacking. In this study, we evaluated the expression of a panel of circulating microRNAs (miRs) involved in the regulation of endothelial function in a population of frail older adults with HFpEF and DM treated for 3 months with empagliflozin, metformin, or insulin. We identified a distinctive pattern of miRs that were significantly regulated in HFpEF patients compared to healthy controls and to HFpEF patients treated with the sodium glucose cotransporter 2 (SGLT2) inhibitor empagliflozin. Three miRs were significantly downregulated (miR-126, miR-342-3p, and miR-638) and two were significantly upregulated (miR-21 and miR-92) in HFpEF patients compared to healthy controls. Strikingly, two of these miRs (miR-21 and miR-92) were significantly reduced in HFpEF patients after the 3-month treatment with empagliflozin, whereas

no significant differences in the profile of endothelial miRs were detected in patients treated with metformin or insulin. Taken together, our findings demonstrate for the first time that specific circulating miRs involved in the regulation of endothelial function are significantly regulated in frail HFpEF patients with DM and in response to SGLT2 inhibition.

SIGNIFICANCE STATEMENT

We have identified a novel microRNA signature functionally involved in the regulation of endothelial function that is significantly regulated in frail patients with HFpEF and diabetes. Moreover, the treatment with the SGLT2 inhibitor empagliflozin caused a modification of some of these microRNAs in a direction that was opposite to what observed in HFpEF patients, indicating a rescue of endothelial function. Our findings are relevant for clinical practice inasmuch as we were able to establish novel biomarkers of disease and response to therapy.

The Santulli's Laboratory is supported in part by the National Institutes of Health National Heart, Lung, and Blood Institute [Grants R01-HL146691, R01-HL164772, R01-HL159062, and T32-HL144456], National Institute of Diabetes and Digestive and Kidney Diseases [Grants R01-DK123259 and R01-DK033823] (to G.S.), by the Diabetes Action Research and Education Foundation (to G.S.), and by the Monique Weill-Caulier and Irma T. Hirschl Trusts (to G.S.). F.V. and J.S.S. are supported by Postdoctoral Fellowships of the American Heart Association [Grants AHA-22POST995561 and AHA-21POST836407], respectively.
dx.doi.org/10.1124/jpet.121.001251.

Introduction

Endothelial dysfunction is a pathogenically relevant mechanism underlying heart failure with preserved ejection fraction (HFpEF) and diabetes mellitus (DM) (Hadi and Suwaidi, 2007; Giamouzis et al., 2016; Gevaert et al., 2019; Knapp et al., 2019; Premer et al., 2019; Jankauskas et al., 2021;

ABBREVIATIONS: BMI, body mass index; BNP, brain natriuretic peptide; CKD, chronic kidney disease; COPD, chronic obstructive pulmonary disease; DBP, diastolic blood pressure; DM, diabetes mellitus; EF, ejection fraction; Empa, empagliflozin; HbA1c, glycated hemoglobin; HDL, high-density lipoprotein; HF, heart failure; HFpEF, heart failure with preserved ejection fraction; Ins, insulin; LDL, low-density lipoprotein; Met, metformin; miR, miRNA (microRNA); SBP, systolic blood pressure; SGLT2, sodium glucose cotransporter 2.

Mone et al., 2021a). HFpEF and DM are very common in older adults, increasing the risk of frailty, a systemic condition that leads to functional decline and adverse outcomes (Owan et al., 2006; Steinberg et al., 2012; Paulus and Tschope, 2013; Chioncel et al., 2017; McHugh et al., 2019; Jankauskas et al., 2021; Lejeune et al., 2021). The pathophysiology of frailty includes chronic inflammation, which is typical of aging (inflammaging), oxidative stress, insulin resistance, loss of anabolic hormones, and reduced tolerance to physical exercise with a reduction in muscle strength (Bandein-Roche et al., 2015; Cruz-Jentoft and Sayer, 2019; Rusanova et al., 2019). Of note, we and others have shown that endothelial dysfunction plays a fundamental role also in the pathobiology of frailty (Alonso-Bouzon et al., 2014; Mansur et al., 2015; Amarasekera et al., 2021; Mone et al., 2021a, 2022a).

Empagliflozin is a relatively novel selective inhibitor of sodium glucose cotransporter 2 (SGLT2) that has been shown to reduce mortality and rehospitalization for HF (Zinman et al., 2015; Anker et al., 2021; Varzideh et al., 2021; Braunwald, 2022). Additional benefits of SGLT2 inhibitors include improved cardiovascular energetics, reduced vascular tone, decreased renal dysfunction, increased circulating levels of ketone bodies, and overall reduced systemic inflammation (Benetti et al., 2016; Prattichizzo et al., 2018; Wan et al., 2018; Oshima et al., 2019; Verma et al., 2019; Zhang et al., 2021; Jensen et al., 2021; Li et al., 2021; Sardu et al., 2021; Varzideh et al., 2021; Huang et al., 2022; Paolisso et al., 2022; Zhang et al., 2022). We have recently demonstrated that empagliflozin significantly improves cognitive impairment in frail older patients with diabetes with HFpEF (Mone et al., 2022c), also showing a correlation between physical and cognitive impairment (Mone et al., 2022a).

MicroRNAs (miRs) are small noncoding RNAs molecules of 18–24 nucleotides, which typically repress mRNAs by binding their 3' untranslated region (Santulli, 2015; Stavast and Erkeland, 2019; Hu et al., 2021; Mirzaei et al., 2021; Mone et al., 2021b; Bielska et al., 2022; Karagiannopoulos et al., 2022; Mauro

et al., 2022; Moisoiu et al., 2022; Qiu et al., 2022; Traber and Yu, 2022; Yaylim et al., 2022; Zeng et al., 2022). Substantial evidence has shown that miRs exert their activity in many biologic processes and several miRs have been proposed as biomarkers and potential targets of novel therapeutic strategies (Creemers et al., 2012; Wronska et al., 2015; Barwari et al., 2016; Zarone et al., 2017; Chen et al., 2018; Wong et al., 2018; Morelli et al., 2019; Kawasaki et al., 2020; Wang et al., 2020; Fonseca et al., 2021; Gambardella et al., 2021; Bonnet et al., 2022; Gambardella et al., 2022a,b; Kansakar et al., 2022; Varzideh et al., 2022). Several investigators have linked miRs to frailty pointing at their involvement in inflammation, endothelial dysfunction, and senescence (Quinn and O'Neill, 2011; Olivieri et al., 2012; Geiger and Dalgaard, 2017; Rusanova et al., 2019; Bu et al., 2021).

In this study, we aimed at assessing the effects of empagliflozin on the profile of circulating miRs involved in the regulation of endothelial function in frail older adults with DM and HFpEF treated with different antidiabetic regimens.

Materials and Methods

Study Design. We evaluated consecutive frail older adults with a confirmed diagnosis of DM and HFpEF, from October 2021 to December 2021. All subjects were recruited from the Sant'Angelo dei Lombardi Hospital, ASL (local health unit of the Italian Ministry of Health) Avellino, Italy. Inclusion criteria were age >65 years; a previous diagnosis of type 2 DM, frailty, and HFpEF; patients were excluded if they had experienced a previous stroke, acute myocardial infarction, or cardiac revascularization. As a control population, we enrolled age-matched subjects with no evidence of HFpEF or DM.

The patients fulfilling the above-mentioned eligibility criteria were divided into three interventional groups (empagliflozin: 10 mg; metformin: 500 mg; and insulin: basal-bolus regimen) and followed-up for three months.

All patients underwent clinical evaluation. Blood samples were taken at baseline and follow up. All patients received a transthoracic echocardiography assessment according to the American Society of Echocardiography recommendations (Lang et al., 2015). Every patient (or a legally authorized representative) signed a written informed

TABLE 1

Baseline characteristics of the patients

Data are means \pm S.D. or *n* (%). "Control" refers to subjects who did not have any evidence of HFpEF or DM.

| | Control | Empagliflozin | Metformin | Insulin |
|-----------------------------|------------------|-------------------|-------------------|-------------------|
| <i>N</i> | 10 | 10 | 10 | 10 |
| Age, y | 79.8 \pm 8.9 | 81.6 \pm 6.8 | 80.8 \pm 6.9 | 81.8 \pm 6.5 |
| Female sex, <i>n</i> (%) | 5 (50.0) | 6 (60.0) | 6 (60.0) | 5 (50.0) |
| BMI (kg/m ²) | 25.6 \pm 1.8 | 27.7 \pm 1.4* | 27.6 \pm 1.7* | 28.1 \pm 1.5* |
| SBP (mmHg) | 118.8 \pm 7.8 | 119.4 \pm 7.2 | 119.8 \pm 7.4 | 120.1 \pm 7.3 |
| DBP (mmHg) | 76.3 \pm 8.8 | 79.0 \pm 7.0 | 79.3 \pm 6.8 | 79.2 \pm 6.9 |
| Heart rate (bpm) | 78.8 \pm 11.1 | 87.3 \pm 8.2 | 86.8 \pm 8.5 | 87.3 \pm 8.6 |
| EF (%) | 65.8 \pm 7.3 | 55.4 \pm 5.2* | 55.8 \pm 5.4* | 55.2 \pm 5.1* |
| Comorbidities, <i>n</i> (%) | | | | |
| Hypertension | 4 (40.0) | 7 (70.0) | 6 (60.0) | 8 (80.0) |
| Dyslipidemia | 7 (70.0) | 8 (80.0) | 8 (80.0) | 7 (70.0) |
| COPD | 4 (40.0) | 4 (40.0) | 5 (50.0) | 6 (60.0) |
| CKD | 3 (30.0) | 5 (50.0) | 6 (60.0) | 7 (70.0) |
| Laboratory parameters | | | | |
| Plasma glucose (mg/dl) | 103.5 \pm 30.6 | 161.8 \pm 39.1* | 163.7 \pm 39.2* | 164.1 \pm 39.0* |
| Cholesterol (mg/dl) | 202.9 \pm 22.1 | 206.1 \pm 20.2 | 205.9 \pm 20.1 | 206.0 \pm 19.8 |
| LDL-cholesterol (mg/dl) | 133.1 \pm 16.1 | 132.3 \pm 19.7 | 132.4 \pm 19.5 | 132.5 \pm 19.8 |
| HDL-cholesterol (mg/dl) | 35.1 \pm 3.5 | 37.5 \pm 3.4 | 36.9 \pm 3.7 | 37.1 \pm 3.4 |
| Creatinine (mg/dl) | 0.9 \pm 0.3 | 1.2 \pm 0.3* | 1.2 \pm 0.4* | 1.3 \pm 0.3* |
| HbA1c (mmol/mol) | — | 56 \pm 6.4 | 55 \pm 7.5 | 57 \pm 5.3 |
| BNP (pg/ml) | — | 443.8 \pm 24.7 | 445.1 \pm 24.5 | 446.2 \pm 25.0 |

BMI, body mass index; BNP, brain natriuretic peptide; CKD, chronic kidney disease; COPD, chronic obstructive pulmonary disease; DBP, diastolic blood pressure; EF, ejection fraction; HbA1c, glycated hemoglobin; HDL, high-density lipoprotein; LDL, low-density lipoprotein; SBP, systolic blood pressure.

**P* < 0.05 versus control.

TABLE 2

Follow up characteristics of the patients 3 months after starting the study

Data are means \pm S.D. or *n* (%). "Control" refers to subjects who did not have any evidence of HFpEF or DM.

| | Control | Empagliflozin | Metformin | Insulin |
|--------------------------|------------------|-------------------|-------------------|-------------------|
| <i>N</i> | 10 | 10 | 10 | 10 |
| BMI (kg/m ²) | 25.4 \pm 1.7 | 27.1 \pm 1.1* | 27.3 \pm 1.2* | 28.0 \pm 1.3* |
| SBP (mmHg) | 117.9 \pm 7.9 | 118.7 \pm 6.8 | 118.6 \pm 6.9 | 120.0 \pm 7.1 |
| DBP (mmHg) | 76.2 \pm 8.7 | 78.9 \pm 6.4 | 79.0 \pm 6.5 | 79.3 \pm 6.8 |
| Heart rate (bpm) | 77.6 \pm 10.3 | 87.0 \pm 7.8* | 86.9 \pm 8.1* | 87.2 \pm 8.2* |
| EF (%) | 65.6 \pm 7.4 | 56.2 \pm 5.0* | 55.9 \pm 5.2* | 55.1 \pm 5.0* |
| Laboratory parameters | | | | |
| Plasma glucose (mg/dl) | 100.2 \pm 28.8 | 159.8 \pm 37.8* | 162.9 \pm 38.6* | 163.3 \pm 38.8* |
| Cholesterol (mg/dl) | 201.5 \pm 22.4 | 205.6 \pm 20.0 | 205.5 \pm 20.3 | 205.9 \pm 19.9 |
| LDL-cholesterol (mg/dl) | 130.1 \pm 16.5 | 131.8 \pm 19.4 | 132.1 \pm 19.3 | 132.3 \pm 19.4 |
| HDL-cholesterol (mg/dl) | 36.1 \pm 3.6 | 37.2 \pm 3.2 | 36.8 \pm 3.6 | 37.0 \pm 3.3 |
| Creatinine (mg/dl) | 0.9 \pm 0.3 | 1.0 \pm 0.2 | 1.0 \pm 0.2 | 1.0 \pm 0.2 |
| BNP (pg/ml) | — | 439.7 \pm 23.8 | 444.5 \pm 24.1 | 444.8 \pm 24.6 |

BMI, body mass index; BNP, brain natriuretic peptide; DBP, diastolic blood pressure; EF, ejection fraction; HDL, high-density lipoprotein; LDL, low-density lipoprotein; SBP, systolic blood pressure.

**P* < 0.05 versus control.

consent. The study was performed in accordance with the ethical standards established in the 1964 Declaration of Helsinki and its later amendments.

Frailty Assessment. A physical frailty assessment was performed following previously described criteria (Mone et al., 2022b,d). A diagnosis of frailty was made with at least three of the following five points: 1) weight loss (unintentional loss of ≥ 4.5 kg in the past year), 2) weakness (handgrip strength in the lowest 20% quintile at baseline, adjusted for sex and body mass index), 3) exhaustion (poor endurance and energy), 4) slowness (walking speed under the lowest quintile adjusted for sex and height), and 5) low physical activity level (lowest quintile of kilocalories of physical activity during the past week).

miR Isolation, Quantification, and Normalization. We extracted miRs using the miRVana miRNA Isolation kit (ThermoFisher) according to the protocol provided by the manufacturer; reverse transcription was performed using the miRCURY LNA Universal RT microRNA PCR kit (Qiagen, Hilden, Germany); miR expression was analyzed by RT-qPCR. We analyzed a panel of miRs that had been previously reported to be involved in the regulation of endothelial dysfunction (Ni et al., 2011; Sabatel et al., 2011; Costa et al., 2013; Zhang et al., 2013; Santulli et al., 2014; Widmer et al., 2014; Kriegel et al., 2015; Ye et al., 2015; Chen et al., 2016; Santulli, 2016; Tang et al., 2017; Cheng et al., 2018; Wei et al., 2018; Gu et al., 2019; Hu and Dong, 2019; Xu et al., 2019; Du et al., 2020; Paterson et al., 2021). The RNA Spike-in kit (Qiagen) was used as an exogenous control of RNA extraction following the manufacturer's instructions. To control yield, we used two synthetic RNA spike-ins (UniSp2 and UniSp5) in different concentrations; miR-320a and miR-423-5p were identified as the most stable miRs among all groups and were therefore used as endogenous normalizers. Relative gene expression was determined using the $2^{-\Delta\Delta CT}$ method.

Statistical Analysis. All data were analyzed using the Prism GraphPad software (Dotmatics, Boston, CA). Data are expressed as means \pm S.D. or numbers and percentages. The differences in miR levels among groups were analyzed using two-tailed *t* tests or one-way ANOVA, followed by Bonferroni post hoc correction, as appropriate.

Results

We enrolled 51 frail older adults with HFpEF and DM. Twenty-one patients were excluded because they did not meet the eligibility criteria, refused to give consent, withdrew from the study, or did not have data from blood analyses at baseline or at follow up. Thus, 30 patients, divided into three treatment groups (empagliflozin, metformin, or insulin)

successfully completed the 3-month follow up. Baseline characteristics of our population are reported in Table 1, whereas follow up data are in Table 2.

Interestingly, the evaluation of the miR signature of endothelial dysfunction revealed a unique pattern of miRs that were significantly regulated in HFpEF patients compared with healthy controls and in HFpEF patients pre and post treatment with the SGLT2 inhibitor empagliflozin (Fig. 1).

We were able to identify three circulating miRs that were significantly downregulated (miR-126, miR-342-3p, and miR-638)

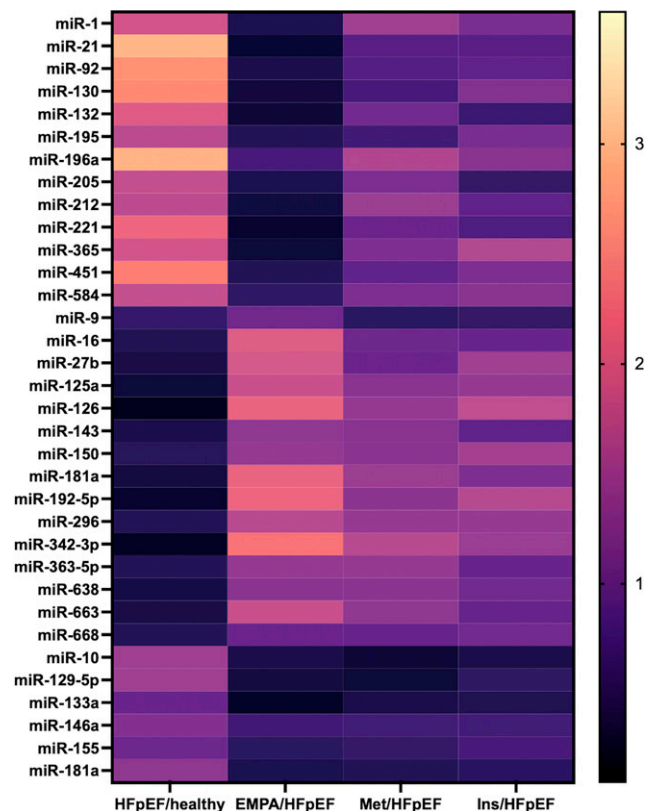


Fig. 1. Heat-map illustrating the expression of circulating miRs in the indicated groups of patients. HFpEF, heart failure with preserved ejection fraction; Healthy, healthy control subjects; Empa, patients receiving empagliflozin; Met, patients receiving metformin; Ins, patients receiving insulin.

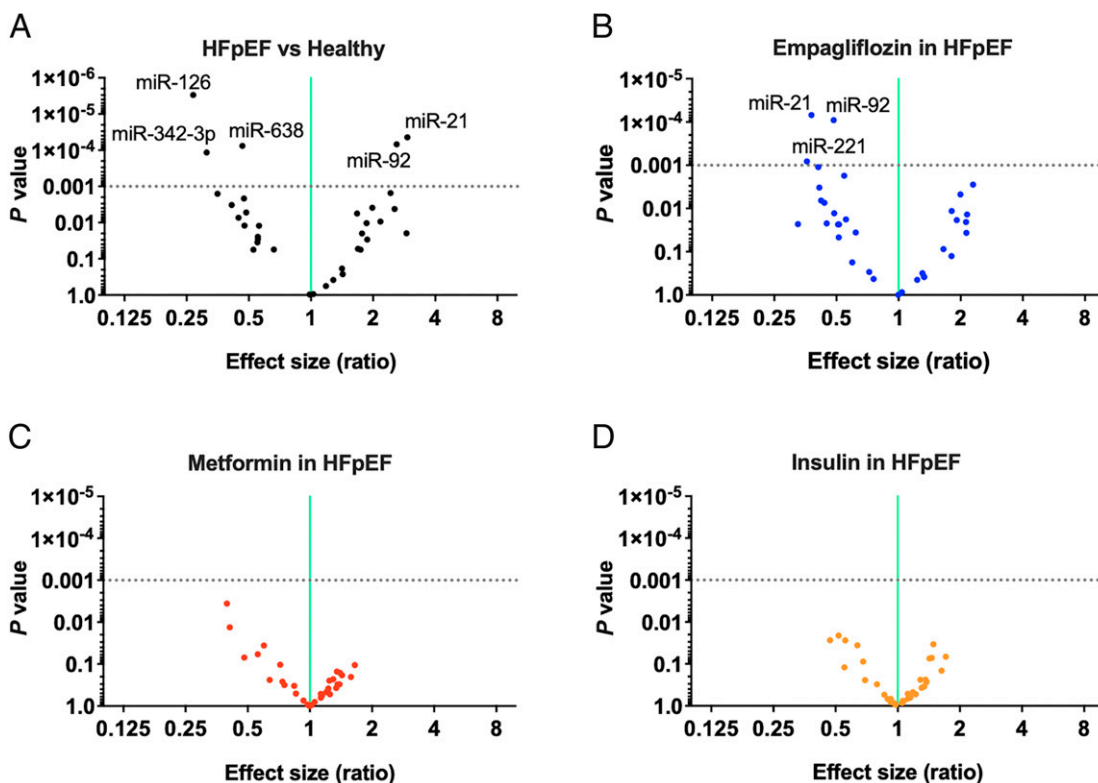


Fig. 2. Volcano plots depicting the miR analyses in the different groups. (A) HFpEF versus healthy controls; (B) effects of empagliflozin treatment in HFpEF patients; (C) effects of metformin treatment in HFpEF patients; and (D) effects of insulin treatment in HFpEF patients. The horizontal dotted line represents a P value of 0.001; thus, the points in the plot above that line represent the differentially expressed miRs with statistical significance.

and two that were significantly upregulated (miR-21 and miR-92) in HFpEF patients compared with healthy controls ($P < 0.001$) (Fig. 2A). Intriguingly, circulating levels of two of these miRs (namely miR-21 and miR-92) were significantly ($P < 0.001$) reduced in HFpEF patients after the 3-month treatment with empagliflozin (Fig. 2B). Instead, no significant differences in the profile of endothelial miRs were detected in patients treated with metformin (Fig. 2C) or insulin (Fig. 2D).

Discussion

To the best of our knowledge, this is the first study investigating the effects of SGLT2 inhibitors on circulating miRs, with a significant relevance both in terms of mechanisms of action and clinical practice. Empagliflozin has been shown to have beneficial effects on cardiovascular outcomes, particularly on the rehospitalization rate for HF (Dave et al., 2020). Nevertheless, there are limited reports investigating the functional role of potential biomarkers to monitor the effects of SGLT2 inhibitors. In this sense, miRs have been widely used as biomarkers; however, limited data are available on the miR profile in frailty (Ipson et al., 2018; Carini et al., 2021). Besides, there are no studies investigating miRs in terms of endothelial dysfunction in HFpEF or frailty.

In our study, we identified five miRs as significantly regulated in HFpEF patients versus healthy control subjects, namely miR-21, miR-92 (upregulated), miR-126, miR-342-3p, and miR-638 (downregulated). Our findings are fully in agreement with previous reports. Indeed, miR-21 has been previously linked to inflammaging and age-related diseases: miR-21

has been proposed as a biomarker of systolic heart failure (Ben-Zvi et al., 2020) and its plasma levels have been linked to aging (Olivieri et al., 2012; Rusanova et al., 2019). Additionally, an increased expression of miR-21 in older adults has been shown to diminish the induction of transcription factor networks involved in memory cell generation (Kim et al., 2018).

Equally important, miR-92 is upregulated after vascular injury, both in vitro and in vivo (Deng et al., 2019), has been previously advocated as a biomarker of HF (Napoli et al., 2020), and its inhibition has been shown to have favorable effects in preventing detrimental cardiac remodeling (Bellera et al., 2014). Strikingly, both miRs were downregulated after empagliflozin treatment, strongly suggesting a rescue of endothelial dysfunction in HFpEF patients after a 3-month treatment with this SGLT2 inhibitor.

Consistent with our data, Cheng and collaborators had demonstrated that miR-342-3p is an indispensable modulator of angiogenic activation in endothelial cells, and downregulation of its expression mediates the vascular dysfunction caused by hyperinsulinemia (Cheng et al., 2018). Further studies are needed to determine the exact clinical relevance of miR-638 downregulation in HFpEF, which could also be compensatory, since previous studies, performed in the setting of hepatocellular carcinoma, suggested that this miR is promoting angiogenesis (Cheng et al., 2016; Yokota et al., 2021).

We observed decreased circulating levels of the master regulator of endothelial function, miR-126 (Liu and Olson, 2010; Santulli et al., 2014; Pei et al., 2020), in HFpEF patients, corroborating the view that endothelial dysfunction is playing an

instrumental role in HFpEF. Consistently, previous analyses had evidenced lower levels of miR-126 in diabetic patients (Zampetaki et al., 2010).

Another miR that was found to be significantly downregulated after empagliflozin treatment is miR-221, which had been linked to muscle proliferation and sarcopenia both in elderly patients and aged mice (Hamrick et al., 2010; He et al., 2020; Roldan Gallardo and Quintar, 2021); the same miR had been also associated with DM and obesity (Lustig et al., 2014). Notably, we did not find evidence of any significant results in terms of endothelial miR network in patients treated with metformin and insulin.

In line with the present findings, most recently we demonstrated that empagliflozin improves endothelial function by reducing mitochondrial calcium overload and the generation of reactive oxygen species (Mone et al., 2022e) and that SGLT2 inhibition has a beneficial impact on quality of life.

In conclusion, our findings demonstrate for the first time that a specific profile of circulating miRs implied in the regulation of endothelial function are significantly regulated in frail HFpEF patients with DM and in response to empagliflozin treatment.

Acknowledgments

The authors thank Dr. X. Wang for insightful discussion.

Author Contributions

Participated in research design: Mone, Lombardi, Frullone, Santulli.

Conducted experiments: Mone, Kansakar, Varzideh, Jankauskas, Pansini, De Gennaro, Famiglietti, Macina, Frullone.

Contributed new reagents or analytic tools: Kansakar, Varzideh, Jankauskas, Pansini, Marzocco, De Gennaro, Famiglietti, Macina, Frullone.

Performed data analysis: Mone, Santulli.

Wrote or contributed to the writing of the manuscript: Mone, Lombardi, Santulli.

References

Alonso-Bouzon C, Carcaillon L, García-García FJ, Amor-Andrés MS, El Assar M, and Rodríguez-Mañas L (2014) Association between endothelial dysfunction and frailty: the Toledo Study for Healthy Aging. *Age (Dordr)* **36**:495–505.

Amarasekera AT, Chang D, Schwarz P, and Tan TC (2021) Does vascular endothelial dysfunction play a role in physical frailty and sarcopenia? A systematic review. *Age Ageing* **50**:725–732.

Anker SD, Butler J, Filippatos G, Ferreira JP, Bocchi E, Böhm M, Brunner-La Rocca HP, Choi DJ, Chopra V, Chuquiure-Valenzuela E, et al.; EMPEROR-Preserved Trial Investigators (2021) Empagliflozin in heart failure with a preserved ejection fraction. *N Engl J Med* **385**:1451–1461.

Bandeian-Roche K, Seplaki CL, Huang J, Buta B, Kalyani RR, Varadhan R, Xue QL, Walston JD, and Kasper JD (2015) Frailty in older adults: a nationally representative profile in the United States. *J Gerontol A Biol Sci Med Sci* **70**:1427–1434.

Barwari T, Joshi A, and Mayr M (2016) MicroRNAs in cardiovascular disease. *J Am Coll Cardiol* **68**:2577–2584.

Bellera N, Barba I, Rodriguez-Sinovas A, Ferret E, Asin MA, Gonzalez-Alujas MT, Pérez-Rodon J, Esteves M, Fonseca C, Toran N, et al. (2014) Single intracoronary injection of encapsulated antagomir-92a promotes angiogenesis and prevents adverse infarct remodeling. *J Am Heart Assoc* **3**:e000946.

Ben-Zvi I, Volinsky N, Grosman-Rimon L, Haviv I, Rozen G, Andria N, Asulin N, Margalit N, Marai I, and Amir O (2020) Cardiac-peripheral transvenous gradients of microRNA expression in systolic heart failure patients. *ESC Heart Fail* **7**:835–843.

Benetti E, Mastrocola R, Vitarelli G, Cutrin JC, Nigro D, Chiazza F, Mayoux E, Collino M, and Fantozzi R (2016) Empagliflozin protects against diet-induced NLRP3 inflammasome activation and lipid accumulation. *J Pharmacol Exp Ther* **359**:45–53.

Bielska A, Niemira M, Bauer W, Sidorowicz I, Szałkowska A, Skwarska A, Raczowska J, Ostrowski D, Gugala K, Dobrzycki S, et al. (2022) Serum miRNA profile in diabetic patients with ischemic heart disease as a promising non-invasive biomarker. *Front Endocrinol (Lausanne)* **13**:888948.

Bonnet H, Bogard B, Hubé F, Ilieva M, Uchida S, Ariza-Mateos MA, Serganov A, Pardini B, Naccarati A, Santulli G, et al. (2022) The non-coding RNA journal club: highlights on recent papers-11. *Noncoding RNA* **8**:31.

Braunwald E (2022) Gliflozins in the management of cardiovascular disease. *N Engl J Med* **386**:2024–2034.

Bu Z, Huang A, Xue M, Li Q, Bai Y, and Xu G (2021) Cognitive frailty as a predictor of adverse outcomes among older adults: a systematic review and meta-analysis. *Brain Behav* **11**:e01926.

Carini G, Musazzi L, Bolzetta F, Cester A, Fiorentini C, Ieraci A, Maggi S, Popoli M, Veronese N, and Barbon A (2021) The potential role of miRNAs in cognitive frailty. *Front Aging Neurosci* **13**:763110.

Chen F, Chen L, He H, Huang W, Zhang R, Li P, Meng Y, and Jiang X (2016) Up-regulation of microRNA-16 in glioblastoma inhibits the function of endothelial cells and tumor angiogenesis by targeting Bmi-1. *Anticancer Agents Med Chem* **16**:609–620.

Chen L, Sun H, Wang C, Yang Y, Zhang M, and Wong G (2018) miRNA arm switching identifies novel tumour biomarkers. *EBioMedicine* **38**:37–46.

Cheng J, Chen Y, Zhao P, Liu X, Dong J, Li J, Huang C, Wu R, and Lv Y (2016) Downregulation of miRNA-638 promotes angiogenesis and growth of hepatocellular carcinoma by targeting VEGF. *Oncotarget* **7**:30702–30711.

Cheng S, Cui Y, Fan L, Mu X, and Hua Y (2018) T2DM inhibition of endothelial miR-342-3p facilitates angiogenic dysfunction via repression of FGF11 signaling. *Biochem Biophys Res Commun* **503**:71–78.

Chioncel O, Lainscak M, Seferovic PM, Anker SD, Crespo-Leiro MG, Harjola VP, Parissis J, Laroche C, Piepoli MF, Fonseca C, et al. (2017) Epidemiology and one-year outcomes in patients with chronic heart failure and preserved, mid-range and reduced ejection fraction: an analysis of the ESC Heart Failure Long-Term Registry. *Eur J Heart Fail* **19**:1574–1585.

Costa A, Afonso J, Osório C, Gomes AL, Caiado F, Valente J, Aguiar SI, Pinto F, Ramirez M, and Dias S (2013) miR-363-5p regulates endothelial cell properties and their communication with hematopoietic precursor cells. *J Hematol Oncol* **6**:87.

Creemers EE, Tjissen AJ, and Pinto YM (2012) Circulating microRNAs: novel biomarkers and extracellular communicators in cardiovascular disease? *Circ Res* **110**:483–495.

Cruz-Jentoft AJ and Sayer AA (2019) Sarcopenia. *Lancet* **393**:2636–2646.

Dave CV, Schneeweiss S, Wexler DJ, Brill G, and Patorno E (2020) Trends in clinical characteristics and prescribing preferences for SGLT2 inhibitors and GLP-1 receptor agonists, 2013–2018. *Diabetes Care* **43**:921–924.

Deng S, Zhang Y, Wang Y, Lu X, and Jiang Q (2019) MicroRNA-92 regulates vascular smooth muscle cell function by targeting KLF4 during vascular restenosis and injury. *Int J Clin Exp Pathol* **12**:4253–4262.

Du X, Hu N, Yu H, Hong L, Ran F, Huang D, Zhou M, Li C, and Li X (2020) miR-150 regulates endothelial progenitor cell differentiation via Akt and promotes thrombus resolution. *Stem Cell Res Ther* **11**:354.

Fonseca A, Ramalhete SV, Mestre A, Pires das Neves R, Marreiros A, Castelo-Branco P, and Roberto VP (2021) Identification of colorectal cancer associated biomarkers: an integrated analysis of miRNA expression. *Aging (Albany NY)* **13**:21991–22029.

Gambardella J, Coppola A, Izzo R, Fiorentino G, Trimarco B, and Santulli G (2021) Role of endothelial miR-24 in COVID-19 cerebrovascular events. *Crit Care* **25**:306.

Gambardella J, Fiordelisi A, Sorriento D, Cerasuolo F, Buonaiuti A, Avvisato R, Pisani A, Varzideh F, Riccio E, Santulli G et al. (2022a) Mitochondrial microRNAs are dysregulated in patients with Fabry disease. *J Pharmacol Exp Ther* DOI: 10.1124/jpet.122.001250 [published ahead of print].

Gambardella J, Kansakar U, Sardu C, Messina V, Jankauskas S, Marfella R, Maggi P, Wang X, Mone P, Paolisso G, et al. (2022b) Exosomal miR-145 and miR-885 Regulate Thrombosis in COVID-19, JPET.

Geiger J and Dalgaard LT (2017) Interplay of mitochondrial metabolism and microRNAs. *Cell Mol Life Sci* **74**:631–646.

Gevaert AB, Boen JRA, Segers VF, and Van Craenenbroeck EM (2019) Heart failure With preserved ejection fraction: a review of cardiac and noncardiac pathophysiology. *Front Physiol* **10**:638.

Giamouzis G, Schelbert EB, and Butler J (2016) Growing evidence linking microvascular dysfunction with heart failure with preserved ejection fraction. *J Am Heart Assoc* **5**:e003259.

Gu X, Wang XQ, Lin MJ, Liang H, Fan SY, Wang L, Yan X, Liu W, and Shen FX (2019) Molecular interplay between microRNA-130a and PTEN in palmitic acid-mediated impaired function of endothelial progenitor cells: effects of metformin. *Int J Mol Med* **43**:2187–2198.

Hadi HA and Suwaidi JA (2007) Endothelial dysfunction in diabetes mellitus. *Vasc Health Risk Manag* **3**:853–876.

Hamrick MW, Herberg S, Arounleut P, He HZ, Shiver A, Qi RQ, Zhou L, Isales CM, and Mi QS (2010) The adipokine leptin increases skeletal muscle mass and significantly alters skeletal muscle miRNA expression profile in aged mice. *Biochem Biophys Res Commun* **400**:379–383.

He N, Zhang YL, Zhang Y, Feng B, Zheng Z, Wang D, Zhang S, Guo Q, and Ye H (2020) Circulating microRNAs in plasma decrease in response to sarcopenia in the elderly. *Front Genet* **11**:167.

Hu C and Dong ZL (2019) MicroRNA-212 promotes the recovery function and vascular regeneration of endothelial progenitor cells in mice with ischemic stroke through inactivation of the notch signaling pathway via downregulating MMP9 expression. *J Cell Physiol* **234**:7090–7103.

Hu L, Wei S, Wu Y, Li S, Zhu P, and Wang X (2021) MicroRNA regulation of the proliferation and apoptosis of Leydig cells in diabetes. *Mol Med* **27**:104.

Huang D, Ju F, Du L, Liu T, Zuo Y, Abbott GW, and Hu Z (2022) Empagliflozin protects against pulmonary ischemia/reperfusion injury via an extracellular signal-regulated kinases 1 and 2-dependent mechanism. *J Pharmacol Exp Ther* **380**:230–241.

Ipsen BR, Fletcher MB, Espinoza SE, and Fisher AL (2018) Identifying exosome-derived microRNAs as candidate biomarkers of frailty. *J Frailty Aging* **7**:100–103.

Jankauskas SS, Kansakar U, Varzideh F, Wilson S, Mone P, Lombardi A, Gambardella J, and Santulli G (2021) Heart failure in diabetes. *Metabolism* **125**:154910.

Jensen J, Omar M, Kistorp C, Tuxen C, Gustafsson I, Køber L, Gustafsson F, Faber J, Malik ME, Fosbøl EL, et al. (2021) Effects of empagliflozin on estimated extracellular

- volume, estimated plasma volume, and measured glomerular filtration rate in patients with heart failure (Empire HF Renal): a prespecified substudy of a double-blind, randomised, placebo-controlled trial. *Lancet Diabetes Endocrinol* **9**:106–116.
- Kansakar U, Varzideh F, Mone P, Jankauskas SS, and Santulli G (2022) Functional role of microRNAs in regulating cardiomyocyte death. *Cells* **11**:983.
- Karagiannopoulos A, Esguerra JLS, Pedersen MG, Wendt A, Prasad RB, and Eliasson L (2022) Human pancreatic islet miRNA-mRNA networks of altered miRNAs due to glycaemic status. *iScience* **25**:103995.
- Kawasaki H, Takeuchi T, Ricciardiello F, Lombardi A, Biganzoli E, Fornili M, De Bortoli D, Mesolella M, Cossu AM, Scrima M, et al. (2020) Definition of miRNA signatures of nodal metastasis in LCa: miR-449a targets notch genes and suppresses cell migration and invasion. *Mol Ther Nucleic Acids* **20**:711–724.
- Kim C, Hu B, Jadhav RR, Jin J, Zhang H, Cavanagh MM, Akondy RS, Ahmed R, Weyand CM, and Goronzy JJ (2018) Activation of miR-21-regulated pathways in immune aging selects against signatures characteristic of memory T cells. *Cell Rep* **25**:2148–2162.e5.
- Knapp M, Tu X, and Wu R (2019) Vascular endothelial dysfunction, a major mediator in diabetic cardiomyopathy. *Acta Pharmacol Sin* **40**:1–8.
- Kriegel AJ, Baker MA, Liu Y, Liu P, Cowley Jr AW, and Liang M (2015) Endogenous microRNAs in human microvascular endothelial cells regulate mRNAs encoded by hypertension-related genes. *Hypertension* **66**:793–799.
- Lang RM, Badano LP, Mor-Avi V, Afilalo J, Armstrong A, Ernande L, Flachskampf FA, Foster E, Goldstein SA, Kuznetsov T, et al. (2015) Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* **28**:1–39.e14.
- Lejeune S, Roy C, Slimani A, Pasquet A, Vancraeynest D, Vanoverschelde JL, Gerber BL, Beaulieu C, and Pouleur AC (2021) Diabetic phenotype and prognosis of patients with heart failure and preserved ejection fraction in a real life cohort. *Cardiovasc Diabetol* **20**:48.
- Li D, Liu Y, Hidru TH, Yang X, Wang Y, Chen C, Li KHC, Tang Y, Wei Y, Tse G, et al. (2021) Protective effects of sodium-glucose transporter 2 inhibitors on atrial fibrillation and Atrial flutter: a systematic review and meta-analysis of randomized placebo-controlled trials. *Front Endocrinol (Lausanne)* **12**:619586.
- Liu N and Olson EN (2010) MicroRNA regulatory networks in cardiovascular development. *Dev Cell* **18**:510–525.
- Lustig Y, Barhod E, Ashwal-Fluss R, Gordin R, Shomron N, Baruch-Umansky K, Hemi R, Karasik A, and Kanety H (2014) RNA-binding protein PTB and microRNA-221 coregulate AdipoR1 translation and adiponectin signaling. *Diabetes* **63**:433–445.
- Mansur HN, Lovisi JC, Colugnati FA, Raposo NR, Fernandes NM, and Bastos MG (2015) Association of frailty with endothelial dysfunction and its possible impact on negative outcomes in Brazilian predialysis patients with chronic kidney disease. *BMC Nephrol* **16**:157.
- Mauro M, Berretta M, Palermo G, Cavalieri V, and La Rocca G (2022) The multiplicity of Argonaute complexes in mammalian cells. *J Pharmacol Exp Ther* DOI: 10.1124/jpet.122.001158 [published ahead of print].
- McHugh K, DeVore AD, Wu J, Matsouka RA, Fonarow GC, Heidenreich PA, Yancy CW, Green JB, Altman N, and Hernandez AF (2019) Heart failure with preserved ejection fraction and diabetes: JACC state-of-the-art review. *J Am Coll Cardiol* **73**:602–611.
- Mirzaei R, Babakhani S, Ajorloo P, Ahmadi RH, Hosseini-Fard SR, Keyvani H, Ahmadyousefi Y, Teimoori A, Zamani F, Karampoor S, et al. (2021) The emerging role of exosomal miRNAs as a diagnostic and therapeutic biomarker in Mycobacterium tuberculosis infection. *Mol Med* **27**:34.
- Moisoù T, Dragomir MP, Iancu SD, Schallenberg S, Birolo G, Ferrero G, Burghlelea D, Stefanuca A, Cozan RG, Licarete E, et al. (2022) Combined miRNA and SERS urine liquid biopsy for the point-of-care diagnosis and molecular stratification of bladder cancer. *Mol Med* **28**:39.
- Mone P, de Donato JA, Varzideh F, Kansakar U, Jankauskas SS, Pansini A, and Santulli G (2022f) Functional role of miR-34a in diabetes and frailty. *Front Aging* **3**:949924 DOI: 10.3389/fragi.2022.949924.
- Mone P, Gambardella J, Lombardi A, Pansini A, De Gennaro S, Leo AL, Famiglietti M, Marro A, Morgante M, Frullone S, et al. (2022a) Correlation of physical and cognitive impairment in diabetic and hypertensive frail older adults. *Cardiovasc Diabetol* **21**:10.
- Mone P, Gambardella J, Pansini A, de Donato A, Martinelli G, Boccalone E, Matarese A, Frullone S, and Santulli G (2021a) Cognitive impairment in frail hypertensive elderly patients: role of hyperglycemia. *Cells* **10**:2115.
- Mone P, Gambardella J, Pansini A, Martinelli G, Minicucci F, Mauro C, and Santulli G (2022b) Cognitive dysfunction correlates with physical impairment in frail patients with acute myocardial infarction. *Aging Clin Exp Res* **34**:49–53.
- Mone P, Gambardella J, Wang X, Jankauskas SS, Matarese A, and Santulli G (2021b) miR-24 targets the transmembrane glycoprotein neuropilin-1 in human brain microvascular endothelial cells. *Noncoding RNA* **7**:9.
- Mone P, Lombardi A, Gambardella J, Pansini A, Macina G, Morgante M, Frullone S, and Santulli G (2022c) Empagliflozin improves cognitive impairment in frail older adults with type 2 diabetes and heart failure with preserved ejection fraction. *Diabetes Care* **45**:1247–1251.
- Mone P, Pansini A, Frullone S, de Donato A, Buonincontri V, De Blasis P, Marro A, Morgante M, De Luca A, and Santulli G (2022d) Physical decline and cognitive impairment in frail hypertensive elders during COVID-19. *Eur J Intern Med* **99**:89–92.
- Mone P, Varzideh F, Jankauskas SS, Pansini A, Lombardi A, Frullone S and Santulli G (2022e) SGLT2 inhibition via empagliflozin improves endothelial function and reduces mitochondrial oxidative stress: insights from frail hypertensive and diabetic patients. *Hypertension* **79**:1633–1643.
- Morelli MB, Shu J, Sardu C, Matarese A, and Santulli G (2019) Cardiosomal microRNAs are essential in post-infarction myofibroblast phenocconversion. *Int J Mol Sci* **21**:201.
- Napoli C, Benincasa G, Donatelli F, and Ambrosio G (2020) Precision medicine in distinct heart failure phenotypes: focus on clinical epigenetics. *Am Heart J* **224**:113–128.
- Ni CW, Qiu H, and Jo H (2011) MicroRNA-663 upregulated by oscillatory shear stress plays a role in inflammatory response of endothelial cells. *Am J Physiol Heart Circ Physiol* **300**:H1762–H1769.
- Olivieri F, Spazzafumo L, Santini G, Lazzarini R, Albertini MC, Rippon MR, Galeazzi R, Abbatecola AM, Marcheselli F, Monti D, et al. (2012) Age-related differences in the expression of circulating microRNAs: miR-21 as a new circulating marker of inflammation. *Mech Ageing Dev* **133**:675–685.
- Oshima H, Miki T, Kuno A, Mizuno M, Sato T, Tanno M, Yano T, Nakata K, Kimura Y, Abe K, et al. (2019) Empagliflozin, an SGLT2 inhibitor, reduced the mortality rate after acute myocardial infarction with modification of cardiac metabolomes and antioxidants in diabetic rats. *J Pharmacol Exp Ther* **368**:524–534.
- Owan TE, Hodge DO, Herges RM, Jacobsen SJ, Roger VL, and Redfield MM (2006) Trends in prevalence and outcome of heart failure with preserved ejection fraction. *N Engl J Med* **355**:251–259.
- Paolisso P, Bergamaschi L, Santulli G, Gallinoro E, Cesaro A, Gragnano F, Sardu C, Mileva N, Foà A, Armillotta M, et al. (2022) Infarct size, inflammatory burden, and admission hyperglycemia in diabetic patients with acute myocardial infarction treated with SGLT2-inhibitors: a multicenter international registry. *Cardiovasc Diabetol* **21**:77.
- Paterson MR, Jackson KL, Dona MSI, Farrugia GE, Visniauskas B, Watson AMD, Johnson C, Prieto MC, Evans RG, Charchar FJ, et al. (2021) Deficiency of microRNA-181a results in transcriptome-wide cell-specific changes in the kidney and increases blood pressure. *Hypertension* **78**:1322–1334.
- Paulus WJ and Tschöpe C (2013) A novel paradigm for heart failure with preserved ejection fraction: comorbidities drive myocardial dysfunction and remodeling through coronary microvascular endothelial inflammation. *J Am Coll Cardiol* **62**:263–271.
- Pei CZ, Liu B, Li YT, Fang L, Zhang Y, Li YG, and Meng S (2020) MicroRNA-126 protects against vascular injury by promoting homing and maintaining stemness of late outgrowth endothelial progenitor cells. *Stem Cell Res Ther* **11**:28.
- Prattichizzo F, De Nigris V, Micheloni S, La Sala L, and Ceriello A (2018) Increases in circulating levels of ketone bodies and cardiovascular protection with SGLT2 inhibitors: is low-grade inflammation the neglected component? *Diabetes Obes Metab* **20**:2515–2522.
- Premer C, Kanelidis AJ, Hare JM, and Schulman IH (2019) Rethinking endothelial dysfunction as a crucial target in fighting heart failure. *Mayo Clin Proc Innov Qual Outcomes* **3**:1–13.
- Qiu JL, Zhang GF, Chai YN, Han XY, Zheng HT, Li XF, Duan F, and Chen LY (2022) Ligustrazine attenuates liver fibrosis by targeting miR-145 mediated TGF-beta/Smad signaling in an animal model of biliary atresia. *J Pharmacol Exp Ther* **381**:257–265.
- Quinn SR and O'Neill LA (2011) A trio of microRNAs that control toll-like receptor signalling. *Int Immunol* **23**:421–425.
- Roldán Gallardo FF and Quintar AA (2021) The pathological growth of the prostate gland in atherogenic contexts. *Exp Gerontol* **148**:111304.
- Rusanova I, Fernández-Martínez J, Fernández-Ortiz M, Aranda-Martínez P, Escames G, García-García FJ, Mañas L, and Acuña-Castroviejo D (2019) Involvement of plasma miRNAs, muscle miRNAs and mitochondrial miRNAs in the pathophysiology of frailty. *Exp Gerontol* **124**:110637.
- Sabatel C, Malvaux L, Bovy N, Deroanne C, Lambert V, Gonzalez ML, Colige A, Rakic JM, Noël A, Martial JA, et al. (2011) MicroRNA-21 exhibits antiangiogenic function by targeting RhoB expression in endothelial cells. *PLoS One* **6**:e16979.
- Santulli G (2015) microRNAs distinctively regulate vascular smooth muscle and endothelial cells: functional implications in angiogenesis, atherosclerosis, and in-stent restenosis. *Adv Exp Med Biol* **887**:53–77.
- Santulli G (2016) MicroRNAs and endothelial (Dys) function. *J Cell Physiol* **231**:1638–1644.
- Santulli G, Wrónska A, Uryu K, Diacovo TG, Gao M, Marx SO, Kitajewski J, Chilton JM, Akat KM, Tuschl T, et al. (2014) A selective microRNA-based strategy inhibits restenosis while preserving endothelial function. *J Clin Invest* **124**:4102–4114.
- Sardu C, Masetti M, Testa N, Martino LD, Castellano G, Turriziani F, Sasso FC, Torella M, De Feo M, Santulli G, et al. (2021) Effects of sodium-glucose transporter 2 inhibitors (SGLT2-I) in patients with ischemic heart disease (IHD) treated by coronary artery bypass grafting via MiECG: inflammatory burden, and clinical outcomes at 5 years of follow-up. *Front Pharmacol* **12**:777083.
- Stavast CJ and Erkeland SJ (2019) The non-canonical aspects of microRNAs: many roads to gene regulation. *Cells* **8**:1465.
- Steinberg BA, Zhao X, Heidenreich PA, Peterson ED, Bhatt DL, Cannon CP, Hernandez AF, and Fonarow GC; Get With the Guidelines Scientific Advisory Committee and Investigators (2012) Trends in patients hospitalized with heart failure and preserved left ventricular ejection fraction: prevalence, therapies, and outcomes. *Circulation* **126**:65–75.
- Tang F, Yang TL, Zhang Z, Li XG, Zhong QQ, Zhao TT, and Gong L (2017) MicroRNA-21 suppresses ox-LDL-induced human aortic endothelial cells injuries in atherosclerosis through enhancement of autophagic flux: involvement in promotion of lysosomal function. *Exp Cell Res* **359**:374–383.
- Traber GM, Yu AM (2022) RNAi based therapeutics and novel RNA bioengineering technologies. *J Pharmacol Exp Ther* DOI: 10.1124/jpet.122.001234 [published ahead of print].
- Varzideh F, Kansakar U, Donkor K, Wilson S, Jankauskas SS, Mone P, Wang X, Lombardi A, and Santulli G (2022) Cardiac remodeling after myocardial infarction: functional contribution of microRNAs to inflammation and fibrosis. *Front Cardiovasc Med* **9**:863238.
- Varzideh F, Kansakar U, and Santulli G (2021) SGLT2 inhibitors in cardiovascular medicine. *Eur Heart J Cardiovasc Pharmacother* **7**:e67–e68.
- Verma S, Mazer CD, Yan AT, Mason T, Garg V, Teoh H, Zuo F, Quan A, Farkouh ME, Fitchett DH, et al. (2019) Effect of empagliflozin on left ventricular mass in patients with type 2 diabetes mellitus and coronary artery disease: the

- EMPA-HEART CardioLink-6 randomized clinical trial. *Circulation* **140**:1693–1702.
- Wan N, Rahman A, Hitomi H, and Nishiyama A (2018) The effects of sodium-glucose cotransporter 2 inhibitors on sympathetic nervous activity. *Front Endocrinol (Lausanne)* **9**:421.
- Wang X, Morelli MB, Matarese A, Sardu C, and Santulli G (2020) Cardiomyocyte-derived exosomal microRNA-92a mediates post-ischemic myofibroblast activation both in vitro and ex vivo. *ESC Heart Fail* **7**:284–288.
- Wei Q, Sun H, Song S, Liu Y, Liu P, Livingston MJ, Wang J, Liang M, Mi QS, Huo Y, et al. (2018) MicroRNA-668 represses MTP18 to preserve mitochondrial dynamics in ischemic acute kidney injury. *J Clin Invest* **128**:5448–5464.
- Widmer RJ, Chung WY, Herrmann J, Jordan KL, Lerman LO, and Lerman A (2014) The association between circulating microRNA levels and coronary endothelial function. *PLoS One* **9**:e109650.
- Wong WKM, Sørensen AE, Joglekar MV, Hardikar AA, and Dalgaard LT (2018) Non-coding RNA in pancreas and β -cell development. *Noncoding RNA* **4**:41.
- Wronska A, Kurkowska-Jastrzebska I, and Santulli G (2015) Application of microRNAs in diagnosis and treatment of cardiovascular disease. *Acta Physiol (Oxf)* **213**:60–83.
- Xu M, Duan Y, and Xiao J (2019) Exercise improves the function of endothelial cells by microRNA. *J Cardiovasc Transl Res* **12**:391–393.
- Yaylim I, Farooqi AA, Telkoparan-Akillilar P, and Saso L (2022) Interplay between non-coding RNAs and NRF2 in different cancers: spotlight on miRNAs and long non-coding RNAs. *J Pharmacol Exp Ther* DOI: 10.1124/jpet.121.000921 [published ahead of print].
- Ye M, Li D, Yang J, Xie J, Yu F, Ma Y, Zhu X, Zhao J, and Lv Z (2015) MicroRNA-130a targets MAP3K12 to modulate diabetic endothelial progenitor cell function. *Cell Physiol Biochem* **36**:712–726.
- Yokota Y, Noda T, Okumura Y, Kobayashi S, Iwagami Y, Yamada D, Tomimaru Y, Akita H, Gotoh K, Takeda Y, et al. (2021) Serum exosomal miR-638 is a prognostic marker of HCC via downregulation of VE-cadherin and ZO-1 of endothelial cells. *Cancer Sci* **112**:1275–1288.
- Zampetaki A, Kiechl S, Drozdov I, Willeit P, Mayr U, Prokopi M, Mayr A, Weger S, Oberholzenzer F, Bonora E, et al. (2010) Plasma microRNA profiling reveals loss of endothelial miR-126 and other microRNAs in type 2 diabetes. *Circ Res* **107**:810–817.
- Zarone MR, Misso G, Grimaldi A, Zappavigna S, Russo M, Amler E, Di Martino MT, Amodio N, Tagliaferri P, Tassone P, et al. (2017) Evidence of novel miR-34a-based therapeutic approaches for multiple myeloma treatment. *Sci Rep* **7**:17949.
- Zeng Q, Qi X, Ma J, Hu F, Wang X, Qin H, Li M, Huang S, Yang Y, Li Y, et al. (2022) Distinct miRNAs associated with various clinical presentations of SARS-CoV-2 infection. *iScience* **25**:104309.
- Zhang A, Luo X, Meng H, Kang J, Qin G, Chen Y, and Zhang X (2021) Sodium glucose cotransporter 2 inhibitors reduce the risk of heart failure hospitalization in patients with type 2 diabetes mellitus: a systematic review and meta-analysis of randomized controlled trials. *Front Endocrinol (Lausanne)* **11**:604250.
- Zhang X, Mao H, Chen JY, Wen S, Li D, Ye M, and Lv Z (2013) Increased expression of microRNA-221 inhibits PAK1 in endothelial progenitor cells and impairs its function via c-Raf/MEK/ERK pathway. *Biochem Biophys Res Commun* **431**:404–408.
- Zhang Y, Liu X, Zhang H, and Wang X (2022) Efficacy and safety of empagliflozin on nonalcoholic fatty liver disease: a systematic review and meta-analysis. *Front Endocrinol (Lausanne)* **13**:836455.
- Zinman B, Wanner C, Lachin JM, Fitchett D, Bluhmki E, Hantel S, Mattheus M, Devins T, Johansen OE, Woerle HJ, et al.; EMPA-REG OUTCOME Investigators (2015) Empagliflozin, cardiovascular outcomes, and mortality in type 2 diabetes. *N Engl J Med* **373**:2117–2128.

Address correspondence to: Dr. Pasquale Mone, 1300 Morris Park Avenue, New York, NY. E-mail: pasquale.mone@einsteinmed.edu
