Endogenous opioids such as enkephalins are activated in response to stress, are implicated in pain control, and mediate systemic and organ-specific responses to injury and adaptation.\(^1,2\) Opioids were initially thought to originate exclusively from the central nervous system, but have since been shown to be expressed in other organs; including the heart, and play a key role in local regulation and response to cardiac injury.\(^1-4\) Their expression in the heart varies in different stages in life, increasing with advanced age and senescence.\(^5,6\) Their effects on myocardial performance are complex. In experimental models, opioid stimulation can result in enhanced myocardial contraction, but can also be accompanied by a transient decrease in heart rate and blood pressure.\(^7\) These effects of bradycardia and hypotension have frequently been characterized as cardiodepressive,\(^1\) but are usually short-lived and transient, and do not reflect long-term hemodynamic or contractile effects.\(^8\)

In cardiac hypertrophy and failure, cardiac opioid system is activated. In experimental models, pressure overload and stimulation of the \(\beta\)-adrenergic signaling pathway leads to increased enkephalin expression\(^8\) suggesting an adaptive and compensatory role to ameliorate the deleterious effects of injury. In studies of patients with heart failure (HF), proenkephalin levels are elevated and are associated with severity of HF and adverse clinical outcomes.\(^9-11\) In a study of 1589 acute and 95 chronic HF patients, elevated proenkephalin levels were predictive of mortality and HF rehospitalizations, but their predictive role was lost with multivariable adjustment when renal markers were entered in the model.\(^10\) Proenkephalin levels were higher in acute, compared with chronic HF patients. Despite the prognostic role of the baseline levels, absolute or relative changes in proenkephalin levels in chronic HF over 6 months, or in acute HF over 7 days during hospitalization, did not provide additional prognostic information, suggesting that the levels are not sensitive to individual patient level changes or response to therapy.\(^10\) In another multicenter study of 1908 patients with acute HF, proenkephalin levels were prognostic of worsening renal function and mortality.\(^9\)

In this issue of *Circulation: Heart Failure*, Emmens et al\(^12\) report that in 2 large independent cohorts of patients with systolic HF, plasma proenkephalin levels were elevated in \(\approx60\%\) of the patients. Levels were higher in older patients, with more severe HF, lower systolic blood pressure, lower rates of HF medica-tions, higher creatinine, lower glomerular filtration rate (GFR), higher natriuretic peptide, and other biomarkers including proinflammatory cytokines.\(^12\) Elevated proenkephalin levels were significantly associated with increased mortality. Initial association with HF hospitalizations became nonsignificant after multivari-able adjustment.
Associations of elevated levels of enkephalins with adverse outcomes in HF raise the question whether sustained expression or overactivation of cardiac enkephalins can be counterregulatory, akin to the maladaptive role of the sustained inflammatory and neurohormonal mediators in chronic HF or whether the elevations are merely a reflection of severity of illness in HF patients, that is, just right amount by the Goldilocks principle for advanced disease state (Figure). In some experimental studies, while opioid receptor agonists reduced reperfusion related release of cardiac injury markers, they promoted reperfusion stunning, and aggravation of posts ischemic systolic and diastolic dysfunction. In salt-sensitive experimental models of hypertension, when administered intravenously, opioid agonists resulted in an immediate decrease in heart rate, an initial small decrease in blood pressure, followed by an increase in blood pressure and subsequent cardiac hypertrophy, suggesting an autocrine/paracrine role for enkephalins in progression of cardiac hypertrophy. Transgenic models of cardiac restricted overexpression of enkephalins do not exist and would be helpful in characterizing their role in experimental HF models. In a murine model of cardiac angiotensin II-overexpression, marked elevation of myocardial enkephalin levels were observed only in the chronic phase when cardiac hypertrophy was apparent at 28 weeks, but not during the early phase, suggesting a delayed and time-dependent role for the activation of the opioid system in the hypertrophied or failing heart, which may be compensatory at the beginning, but could be maladaptive, if not turned off. Thus, the timing of these mediators is critical in defining their adaptive versus maladaptive role in HF. Furthermore, cardioprotective opioidergic responses may be blunted or desensitized with aging or chronic disease states such as HF, and increased levels of enkephalins may reflect an adaptive response to attenuation of opioid responsiveness. Hence, elevations in circulating levels of enkephalins likely represent advanced disease state, and are associated with adverse prognosis and clinical outcomes, similar to the prognostic role of natriuretic peptides in HF. Given their differing roles ranging from adaptive, compensatory to potentially unfavorable at different stages of cardiac failure, elevated levels of enkephalins alone are not adequate to imply causality, or justify their characterization as biological markers causing or worsening HF.

Regarding their renal effects, in animal studies, while certain opioid agonists have been shown to stimulate diuresis and natriuresis, others have been reported to
exert antidiuretic effects via antidiuretic hormone secretion.\textsuperscript{15,16} Interestingly, even for the same agonist, diuretic effects can change depending on volume status.\textsuperscript{16} In the study by Emmens et al\textsuperscript{12} deterioration of kidney function, defined as >25% decrease in estimated GFR over 9 months, was seen in 22% of the patients, and was predicted by doubling of proenkephalin levels. Because of its small molecular mass and absence of a binding protein, proenkephalin is thought to be freely filtrated through the glomerulus and therefore a good measure of GFR.\textsuperscript{10} In the study by Emmens et al,\textsuperscript{12} in addition to glomerular dysfunction, proenkephalin levels were also strongly associated with renal tubular markers. In another study of patients with acute HF, though proenkephalin levels strongly correlated with renal blood flow and GFR, they were not associated with renal tubular markers.\textsuperscript{10} In other studies of patients with acute kidney injury markers after cardiac surgery, or myocardial infarction, proenkephalin levels were shown to rapidly increase.\textsuperscript{10} Hence, the role of enkephalins in the kidney appears to be complex: proenkephalin can be a measure of GFR, a marker of kidney injury in acute injury states such as myocardial infarction or cardiac surgery, and a marker of advanced disease state in chronic HF. It is unclear whether enkephalins have a mechanistic role for development or prevention of kidney injury, and thus remain with an unclear role as markers of disease severity affecting kidney function such as HF, or a biological marker involved in pathogenesis of kidney injury.

Furthermore, proenkephalin levels can vary according to patient characteristics and can be challenging to measure as they require sandwich immunoassay measurements. Proenkephalin levels can rise with age, comorbidities, and other injuries. The normal range reported in a general population is quite wide, between 9 and 518 pmol/L,\textsuperscript{12} raising the question whether a median of 45 pmol/L or 99th percentile cutoff 80 pmol/L would be an appropriate relative reference value for an older HF population with comorbidities. In the study by Emmens et al,\textsuperscript{12} the levels were elevated in sicker and older patients, and residual confounding despite multivariable adjustment likely played a role for the association of proenkephalin levels with adverse outcomes.

It is also important to recognize that treatment of HF can affect the enkephalin levels. ACE (angiotensin-converting enzyme) and neprilysin break down encephalin—interestingly, neprilysin originally was named as enkephalinase in early 1970s.\textsuperscript{17} Therefore, treatment with ACE inhibitors or sacubitril/valsartan, an ARNi (angiotensin receptor antagonist with neprilysin inhibitor), can result in inhibition of degradation of enkephalin, and a rise in enkephalin levels in patients with HF. It remains to be elucidated whether such potential increases in enkephalin levels can explain or potentiate the beneficial effects of ACE inhibitor or ARNi, or could be maladaptive in patients with HF.

The questions of whether the proenkephalin levels in HF patients are markers of disease severity, versus markers of maladaptive counterregulation by an over-activated opioid system remain unanswered.

Until better experimental and clinical characterization of the acute and long-term cardiovascular effects of intrinsic opioid system are available, it would be very difficult to define the right timing or the right levels of enkephalins in HF. Until then, proenkephalin joins the multitude of other markers suffering from mistaken causation and the repeated epithets of Goldilocks syndrome; implicated for worse prognosis in patients with advanced HF, but without a clear characterization of their potential double-edged role of early cardioprotection versus late maladaptation, and without an elucidation on how much is too much versus just right (Figure) at individual patient levels, at different times.

REFERENCES

1. van den Brink OW, Delbridge LM, Rosenfeldt FL, Penny D, Esmore DS, Quick D, Kaye DM, Pepe S. Endogenous cardiac opioids: enkephalins in adaptation and protection of the heart. Heart Lung Circ. 2003;12:178–187. doi: 10.1016/j.hlc.2003.01.006
2. Headrick JP, See Hoe LE, Du Toit EF, Peart JN. Opioid receptors and cardioprotection - ‘opioidergic conditioning’ of the heart. Br J Pharmacol. 2015;172:2026–2050. doi: 10.1111/bph.13042
3. Pugsley MK. The diverse molecular mechanisms responsible for the actions of opioids on the cardiovascular system. Pharmacol Ther. 2002;93:51–75.
4. Nguyen VT, Wu Y, Guillery AN, McConnell BK, Fujise K, Huang MH. Delta-opioid augments cardiac contraction through [beta]-adrenergic and CGRP-receptor co-signaling. Peptides. 2012;33:77–82. doi: 10.1016/j.peptides.2011.11.010
5. Caffrey JL, Boluyt MO, Younes A, Barron BA, O’Neill L, Crow MT, Lakatta EG. Aging, cardiac proenkephalin mRNA and enkephalin peptides in the fisheries 344 rat. J Mol Cell Cardiol. 1994;26:701–711. doi: 10.1006/jmcc.1994.1085
6. van den Brink OW, Delbridge LM, Pedrazzini T, Rosenfeldt FL, Pepe S. Augmented myocardial methionine-enkephalin in a murine model of cardiac angiotensin II-overexpression. J Renin Angiotensin Aldosterone Syst. 2007;8:153–159. doi: 10.3317/jraas.2007.030
7. Fujita S, Smart SC, Stowe DF. Enhanced contractile responsiveness to cytosolic Ca(2+) by delta-2 opioid agonist deltorphin in intact guinea pig hearts. J Mol Cell Cardiol. 2000;32:1647–1659. doi: 10.1006/jmcc.2000.1199
8. Weil J, Zolk O, Grienertog J, Wenzel U, Zimmermann WH, Eschenhagen T. Alterations of the proenkephalin system in cardiac hypertrophy and

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Disclosures

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its role in atrioventricular conduction. Cardiovasc Res. 2006;69:412–422. doi: 10.1016/j.cardiores.2005.10.016

9. Ng LL, Squire IB, Jones DJL, Cao TH, Chan DCS, Sandhu JK, Quinn PA, Davies JE, Struck J, Hartmann O, Bergmann A, Mebazaa A, Gayat E, Arrigo M, Akiyama E, Sabti Z, Lohmann J, Twerenbold R, Herrmann T, Schumacher C, Kozhuharov N, Mueller C; GREAT Network. Proenkephalin, renal dysfunction, and prognosis in patients with acute heart failure: a great network study. J Am Coll Cardiol. 2017;69:56–69. doi: 10.1016/j.jacc.2016.10.038

10. Matsue Y, Ter Maaten JM, Struck J, Metra M, O’Connor CM, Ponikowski P, Teerlink JR, Cotter G, Davison B, Cleland JG, Givertz MM, Bloomfield DM, Dittrich HC, van Veldhuisen DJ, van der Meer P, Damman K, Voors AA. Clinical correlates and prognostic value of proenkephalin in acute and chronic heart failure. J Card Fail. 2017;23:231–239. doi: 10.1016/j.cardfail.2016.09.007

11. Kawashima S, Fukutake N, Nishian K, Asakuma S, Iwasaki T. Elevated plasma beta-endorphin levels in patients with congestive heart failure. J Am Coll Cardiol. 1991;17:53–58.

12. Emmens JE, ter Maaten JM, Damman K, van Veldhuisen DJ, de Boer RA, Struck J, Bergmann A, Samaîî E, Streng KW, Anker SD, Dickstein K, Lang CC, Metra M, Samani NJ, Ng LL, Voors AA. Proenkephalin, an opioid system surrogate, as a novel comprehensive renal marker in heart failure. Circ Heart Fail. 2019;12:e005544. doi: 10.1161/CIRCHEARTFAILURE.118.005544

13. Lasukova TV, Maslov LN, Gorbunov AS. Effects of μ-opioid receptor agonist remifentanil on heart contractility and necrotic injury to cardiomyocytes during ischemia and reperfusion of isolated rat heart. Bull Exp Biol Med. 2015;159:722–725. doi: 10.1007/s10517-015-3058-7

14. Hao JM, Rabkin SW. Increased cardiac ppENK mRNA in cardiac hypertrophy and effects on blood pressure of its peptide products. Am J Physiol. 1997;272(pt 2):H2885–H2894. doi: 10.1152/ajpheart.1997.272.6.H2885

15. Sezen SF, Kenigs VA, Kapusta DR. Renal excretory responses produced by the delta opioid agonist, BW373U86, in conscious rats. J Pharmacol Exp Ther. 1998;287:238–245.

16. Kapusta DR. Opioid mechanisms controlling renal function. Clin Exp Pharmacol Physiol. 1995;22:891–902.

17. Skidgel RA, Erdos EG. Angiotensin converting enzyme (ACE) and nephrisin hydrolyze neuropeptides: a brief history, the beginning and follow-ups to early studies. Peptides. 2004;25:521–525. doi: 10.1016/j.peptides.2003.12.010