

Review

Sex beyond cardiovascular risk factors and clinical biomarkers of cardiovascular diseaseMaria Bergami¹, Marialuisa Scarpone¹, Raffaele Bugiardini¹, Edina Cenko¹,
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Abstract

In recent years, increasing attention has been reserved to the analysis of sex-related differences in pathophysiology and prognosis of ischemic heart disease (IHD). The traditional conventional cardiovascular risk factors (hypertension, hypercholesterolemia, diabetes mellitus and cigarette smoking) are still considered the major risk factors for IHD in both sexes. Nevertheless, recent studies show that they may interact with male and female coronary anatomy in a different manner. The path to sex-specific risk stratification of IHD is also supported by differences in inflammation and necrosis biomarkers (such as C-reactive protein and troponins, respectively). Indeed, large cohort studies often show different mean values of these markers in men and women. The current review summarizes the state-of-art knowledge on sex-related differences in cardiovascular risk factors and cardiac biomarkers with a prognostic value.

Keywords: Ischemic heart disease; Sex differences; Women; Biomarkers; Risk factors**1. Sex differences in myocardial infarction and ischemia**

Over the past decades, increasing attention has been reserved to the analysis of differences between men and women in the pathophysiology, diagnostic, and prognosis of cardiovascular diseases (CVD) [1–5]. This issue first acquired relevance in the early 90s, when it became apparent that the majority of randomized control trials (RCTs) excluded or included only a strict minority of women in their cohorts [6]. This phenomenon was partly based on the assumption that men are more frequently victims of CVD than women, and that the medications administered to men would automatically work, in the same proportion and with the same dosages, in the few women affected by the disease. These assumptions have been proven over time to be misconceptions. In fact, while it is true that women develop CVD approximately 10 years later than men, it has also been observed that female patients do have higher early case fatality rates and higher 1-year mortality rates, when affected by myocardial infarction [7–9]. The reasons behind this phenomenon are complex [10], and are still subject to studies and debates.

Gender and sex issues have been the main focus of research in the last years. Despite being considered similar enough to men to not being included in RCTs, women have been victim of undertreatment and underdiagnosis in many medical fields, especially in CVD. Focusing on ischemic heart disease (IHD) women have been reported to have longer delays from symptom onset to hospital presentation than men [11,12]. In addition, women more commonly have symptoms such nausea, palpitations, and short-

ness of breath, rather than chest pain, thus when they arrive at the hospital, the atypical nature of their symptoms often proves to be misleading for physicians, bringing to further delays in the diagnosis of acute coronary syndromes (ACS). Moreover, even after a timely diagnosis, women do not always receive the same level of care as men do, as demonstrated by the lower rates of both medical and invasive treatment described in the female sex by several authors [11,13].

Despite being undoubtedly worrying in their nature and extent, gender issues do not appear to be the sole cause of the differences between women and men in the survival and quality of life after an ACS. In a recent study by Cenko *et al.* [14] it was demonstrated that female patients, especially if younger than 60 years, have higher rates of 30-day mortality after ST-segment elevation myocardial infarction (STEMI) than younger men did even after adjustment for comorbidities and treatment covariates (odd ratio [OR], 1.88; 95% confidence interval [CI], 1.04–3.26). These data, supported by findings from other cohorts, clearly suggest that the differences in prognosis after an acute myocardial infarction (AMI) in men and women cannot be reconducted to gender-related disparities alone, but may be caused by intrinsic biological differences between the two sexes. Evidence supporting this hypothesis can be found in the pathophysiology of IHD. Women appear to present sex specific mechanisms underlying the clinical manifestations of IHD. Several studies showed that women have a less plaque burden than men, even despite the presence of higher rates of cardiovascular risk factors and even in the early stages of coronary atherosclerosis [15,16]. In addition, women have more diffuse epicardial endothelial dysfunction than men.



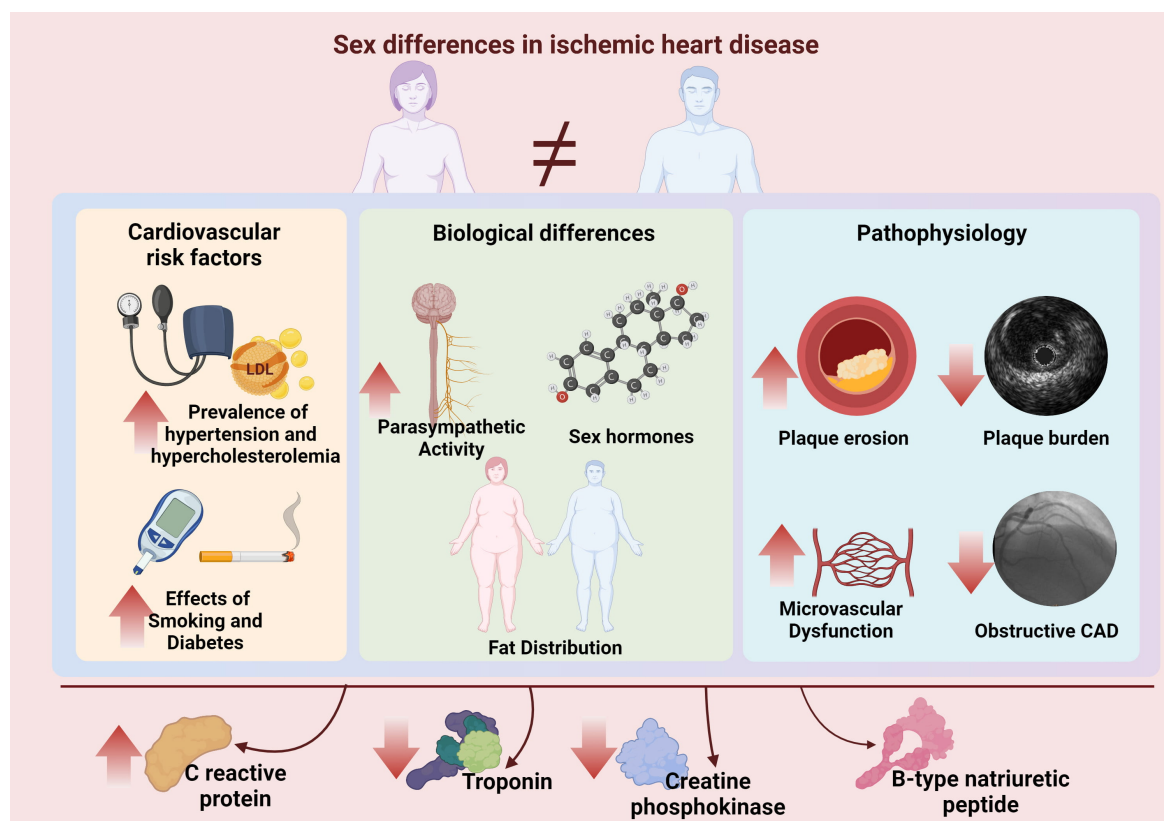


Fig. 1. Main differences in ischemic heart disease between men and women. Women have a higher prevalence of hypertension and hypercholesterolemia, but lower levels of abdominal obesity, lower plaque burden and obstructive coronary artery disease. Ischemic heart disease in women occurs with plaque erosion and microvascular dysfunction more often than men. The higher vagal tone and estrogen levels (in the premenopausal state) postpone the development of the coronary artery disease by approximately 8 years compared with men. Nevertheless, women have worse outcomes, especially when they are smokers or affected by diabetes mellitus. All these differences may be contributing factors to the different expression of cardiovascular biomarkers between sexes.

Also, there are important sex differences in the autonomic nervous control of the cardiovascular system are described as well. Men tend to have a higher sympathetic cardiac autonomic activity, whereas women tend to have a higher parasympathetic activity [17] (Fig. 1). Plaque erosion appears to be the most frequent substrate to the acute coronary event in women, whereas in men plaque rupture is more likely [18,19]. This might impact the distribution of the types of IHD across sexes: female patients have lower rates of STEMI, but higher rates of non-ST segment elevation acute coronary syndromes (NSTEMI) and chronic coronary syndromes. Yet, in case of STEMI, women have a higher likelihood of developing severe complications like acute heart failure, with detrimental effects on prognosis [20].

The evidence of biological differences between sexes has consequences not only on the therapeutic strategies and medications administered to women with IHD, but also on the approaches used to evaluate patients' risk of developing the disease or even diagnose IHD in the acute phase and its prognosis.

Sex differences in the role of cardiovascular risk fac-

tors are prone to translate into discrepancies in pathophysiologic mechanisms underlying myocardial ischemia and expression of serum CVD biomarkers between sexes (Fig. 1). The current review aims to report current evidence on these aspects.

2. Cardiovascular risk factors

Hypertension, hypercholesterolemia, diabetes mellitus and cigarette smoking are the major cardiovascular factors for the development of significant coronary artery disease (CAD) in the general population. However, there are only few data regarding the existence of any sex differences in the impact of these risk factors and how these differences could affect the severity of CAD or its relationship to outcomes. Huxley *et al.* [21] has suggested that smoking has a much larger harmful impact on women. Other studies have shown that smoking has a similar effect on increasing the risk of CHD in both men and women [22]. Recently, data coming from the ISACS-TC (International Survey of Acute Coronary Syndromes in Transitional Countries) shed light on the topic [23]. The study demonstrated that the harms of smoking differ by sex. Compared with nonsmok-

ers, women (but not men) who were current smokers had a much greater risk of ACS with obstructive CAD than non-obstructive CAD. The study also showed that hypercholesterolemia and hypertension increased the risk of obstructive CAD without any differences between men and women, although the latter has a remarkably higher prevalence in the female sex [23]. These data are in line with other observational studies [23]. Moreover, hypertension particularly affects the coronary microcirculation, which therefore becomes extremely vulnerable in postmenopausal women, due to the drop in estrogen which plays a protective role [19,24]. Many studies describe that women with diabetes mellitus face an increased cardiovascular risk compared to men [22,23].

Obesity is a strong predictor of overall mortality, and especially abdominal obesity [25]. However, it seems that there is a significant interaction between body mass index (BMI) and sex in regard to the risk of AMI and cardiovascular death. Indeed, obese men have an increased risk of AMI and cardiovascular death, but, surprisingly, overweight women had a decreased risk of AMI compared with normal weighted women [25,26].

Sedentary life is another risk factor for IHD. Several studies demonstrated that physical activity has a protective role both in primary and secondary prevention for men and women. Exercise training interventions have vasoprotective and antiatherosclerotic effects, likely due to the decrease of body fat, and the improvement of hypertension, dyslipidemia and diabetes. However, to enhance the complexity of interaction between sex and risk of IHD, no sex differences have been reported in the positive response of exercise training [27].

In conclusion, smoking and diabetes mellitus disproportionately increase the risk of obstructive CAD in women, and as so they are key factors in explaining sex differences in outcomes from ACS. Intense efforts to reduce tobacco use and increase screening for pre-diabetes mellitus have the potential to reduce the sex lag in cardiovascular disease mortality in women compared with men. Moreover, women would benefit more from strategies that prevent and treat hypertension at the population level and not at the individual one.

3. Markers of inflammation

Early and accurate CVD risk prediction and the subsequent implementation of prevention strategies determine a clear beneficial impact on patients' health and quality of life, independently from sex. Considerable effort has been placed upon improving cardiovascular risk prediction with the use of biomarkers that play a key role in the pathogenesis of the disease or reflect atherosclerotic status. Still today, however, the most frequently discussed biomarkers in the primary prevention setting remain those index of systemic inflammation [28].

Systemic inflammation has been extensively demonstrated to be strictly related to the patient's metabolic state and lipid profile. Increased levels of inflammation markers have also been associated with a higher risk of developing CVD, especially in women [29,30].

One of the most extensively studied markers of inflammation in cardiology is C-reactive protein (CRP), a protein synthesized by the liver whose clinical use in cardiovascular prevention has been endorsed either by the current American Heart Association/American College of Cardiology (AHA/ACC) and European Society of Cardiology (ESC) guidelines, especially in regards to its high sensitivity quantification [28,31,32]. Current recommendations for CRP testing suggest uniform CRP thresholds to characterize the relative risk of CV events, but both sex and race-related differences in the expression of this biomarker have been reported [33,34].

In a study by Khera *et al.* [32], female sex and black race were both associated with higher CRP levels (median, 3.3 vs. 1.8 mg/L in women versus men, and 3.0 vs. 2.3 mg/L in Caucasian vs. Black patients; $p = 0.001$). Sex difference was reported to be independent from the use of statins or oral estrogens, and was only partially attenuated by adjustment for traditional CV risk factors, BMI, statin, estrogen use, and sampling weights (OR, 1.6; 95% CI, 1.1–2.5 and OR, 1.7; 95% CI, 1.2–2.6, respectively). These findings were confirmed by a more recent analysis by Lau *et al.* [35], conducted on 7184 patients from the Framingham Heart Study. The authors observed marked differences in expression of biomarkers representing pathways of inflammation, including CRP. Interestingly, differently from what suggested in previous studies [36,37], sex-related differences were not influenced by women's menopausal state. These data suggest the potential usefulness of this biomarker in the evaluation of cardiovascular risk and in the stratification and personalization of said estimates based on sex. Such a possibility is further endorsed by the different relationship of CRP with the patient's metabolic state that was observed across sexes. In fact, while other authors confirmed the persistence of sex differences in CRP levels independently from BMI [33], women with higher BMI were reported to have higher CRP levels than normal weighted women, whereas the phenomenon was not observed in men [32]. Table 1 (Ref. [32,33,38–41]) reports some large observational studies exploring sex differences in CRP levels in individuals without history of IHD. Moreover, the use of alternative measures for patient's weight status, such as distribution of body fat, suggested that the presence of increased subcutaneous adiposity was associated with higher CRP levels in women, but not in men [41]. Similar discrepancies were observed also in individuals with diabetes and metabolic syndrome [42,43].

Table 1. Sex differences in C-reactive protein on large observational studies with individuals with no history of IHD.

| Authors | Registry/Country | Year | Population | Women | Men | <i>p</i> value | Note |
|------------------------------|---|-----------|------------|---------------------|---------------------|----------------|--|
| | | | | (mg/L) | (mg/L) | | |
| Rutter <i>et al.</i> [39] | Framingham Offspring Study | 1991–1995 | 3037 | 1.6 (0.2–5.1) | 1.3 (0.3–4.0) | 0.002 | Highest values in women with DM or MetS |
| Khera <i>et al.</i> [32] | Dallas Heart Study | 2005 | 6101 | 3.3 | 1.8 | 0.001 | Strong association with BMI |
| Rogowski <i>et al.</i> [33] | Israel | 2004 | 938 | 2.1 ± 3.4 | 1.5 ± 2.8 | <0.0005 | Subjects with CV risk factors |
| Rogowski <i>et al.</i> [33] | Israel | 2004 | 410 | 1.6 ± 3.4 | 1.0 ± 2.7 | <0.0005 | Subjects with no CV risk factors |
| Cartier A <i>et al.</i> [41] | Canada | 2009 | 353 | 1.24 (0.54–3.04) | 0.94 (0.51–2.40) | <0.05 | Premenopausal women |
| Metha NN <i>et al.</i> [40] | Penn Diabetes Heart Study | 2011 | 1299 | 2.8 (1.4–6.6) | 1.4 (0.7–2.9) | <0.001 | Diabetic subjects |
| Metha NN <i>et al.</i> [40] | Study of Inherited Risk of Coronary Atherosclerosis | 2011 | 860 | 1.4 (0.6–3.7) | 1.1 (0.5–2.1) | <0.001 | Nondiabetic subjects |
| Lakoski <i>et al.</i> [38] | Multiethnic Study of Atherosclerosis | 2006 | 6814 | 2.56 (4.6) | 1.43 (2.5) | 0.0001 | Women on estrogen: median 3.90 Women not on estrogen: median 2.15 |

Data are mean ± SD or median (interquartile range).

CV, cardiovascular; DM, diabetes mellitus; BMI, body mass index; MetS, Metabolic Syndrome.

4. Markers of cytonecrosis

4.1 Troponins

Since these proteins are part of the contractile system of the myocardium, increases in levels of cardiac troponin I (cTnI) and T (cTnT) are quite specific indicators of cardiac injuries. For this reason, cardiac troponins and high-sensitivity cardiac troponins (hs-Tn) are the markers of myocardial injury most widely used in clinical practice and endorsed by current Guidelines on management and treatment of myocardial ischemia [44].

According to the Fourth Universal Definition of Myocardial Infarction, myocardial injury is defined as elevation of cTn values above the 99th percentile upper reference limit, with dynamic changes in level of this biomarker being indicative of an acute injury [44]. Still, there have been controversies regarding the optimal threshold to apply to each assay. In fact, baseline levels of cTn and hs-cTn have been demonstrated to be highly dependent on age, kidney injury, blood flow, timing of blood samples and biological variation. Sex differences in levels of this biomarker have also been observed, although the clinical importance of these differences has been subject to debate [45].

Either in animal and human models, women appear to present with lower levels of cTn levels than men do [42]. In a Treat Angina with Aggrastat and determine Cost of Therapy with an Invasive or Conservative Strategy (TACTICS-TIMI 18) substudy, Wiviott *et al.* [46] observed that, after adjustment for demographic characteristics, risk factors and history of CVD, women were less likely to have positive TnT than their male counterparts (OR, 0.53; 95% CI, 0.43–0.68). This has been attributed to intrinsic differences between the sexes: aside from the aforementioned differences in CAD extension (Fig. 1), which may bring to consequent

discrepancies in the release of markers of cytonecrosis, troponin concentrations correlate with left ventricular mass, which is typically greater in men [45,47].

In the TACTICS-TIMI 18 substudy, the presence of elevated troponins in female sex was also associated with higher recurrence rates of myocardial infarction at 180 days (OR for TnI, 0.30; 95% CI, 0.10–0.86). Eggers *et al.* [48] confirmed the previously observed sex-related difference in expression of this biomarker, as in their analysis median levels of cTnI were 4.1 (2.9–6.5) ng/L in men and 3.0 (2.3–3.9) ng/L in women ($p < 0.001$), but terms for sex interaction on the association between cTnI levels and all-cause mortality and incident CVD were not significant in a median follow-up of 10 years.

Despite these evidences, the suboptimal analytical precision at low concentrations of cTn does not allow the definition of sex-specific cTn 99th percentiles for conventional assays. This issue has been partly solved with the development of high-sensitivity assays, which have further highlighted the aforementioned sex-related discrepancies [45]. In this regard, in a cross-sectional analysis performed in three independent community-based cohorts, Gore *et al.* [49] observed higher 99th percentile cutpoints of hs-cTnT in men compared with women, with median values ranging from 17 to 39 ng/L and from 11 to 34 ng/L in men versus women, respectively, depending from the cohort and sub-populations considered. Shah *et al.* [47] provided further insight into this phenomenon. In their study, the use of a high sensitivity assay (Abbott), significantly increased the number of diagnoses of type I myocardial infarction in women, especially if a sex-specific threshold was applied. This increase was observed only marginally in men. The use of a sex-specific threshold (34 ng/L in men and 16 ng/L in women) proved also to be a more accurate predictor of

recurrent myocardial infarction in women, suggesting that acknowledging these differences in biomarkers expression would prove to be beneficial in clinical practice, especially in terms of diagnostic accuracy for female patients. Following these observations, and despite the presence of dissenting voices regarding the clinical impact of different hs-cTn thresholds in men and women [50], the identification of sex-specific cutoffs for hs-cTn assays has been suggested by the Global Task Force for the Diagnosis of MI and the IFCC (International Federation of Clinical Chemistry and Laboratory Medicine) Task Force on Cardiac Biomarkers [51]. Still, while the most recent ESC Guidelines for management of ACS without persistent ST segment elevation did provide assay-specific cut-offs for hs-Tn evaluation according to the 0/1 h or 0/2 h algorithms, they did not differentiate these thresholds by sex. Indeed, it was deemed necessary to wait for the development of a stratification tool that could take into consideration also patient's age, renal function and time of chest pain onset [52].

4.2 Other biomarkers of cytonecrosis

Aside from cardiac troponins, among the remaining biomarkers of myocardial cytonecrosis those with the highest clinical impact are creatine phosphokinase (CK) and copeptin, although they all present levels of specificity and sensitivity markedly inferior to cTn [52].

CK is an enzyme apt at promoting the transfer of high-energy phosphate into and out of mitochondria. The isoenzyme CK-MB (combination of brain and muscle subunits) is expressed in high concentrations in the myocardium. Consequently, myocardial injury of any type usually leads to an increase in CK-MB plasma levels. Dosing this biomarker has been for decades an essential aid in the diagnosis of myocardial infarction, right before cTn assays were introduced to clinical practice. Still, as CK-MB shows a more rapid decline after myocardial infarction than cTn, its use is still endorsed as further evidence to establish the timing of myocardial injury and detection of early infarction. CK-MB is considered the best alternative to cTn in case of unavailability of the latter [44,52]. As even in this case women tend to present lower serum level of CK-MB than men do, sex-specific thresholds are available and recommended for use [44].

In regards to copeptin, a neuropeptide of yet undetermined pathophysiological function, its use is emerging as an additional biomarker for rapid rule-out of myocardial infarction in case of absence of hs-cTn assays [52]. Data regarding potential sex differences in the expression of this biomarker are scarce, although in a recent analysis by Vargas *et al.* [53] male sex was associated with higher levels of this biomarker (OR, 2.37; 95% CI, 1.61–3.49; $p < 0.001$).

5. Markers of myocardial dysfunction

Acute heart failure is one of the most common and dangerous complications of myocardial ischemia, with a

deep impact on a patient's prognosis and quality of life. The most renowned biomarkers for this condition are B-type natriuretic peptide (BNP) and N-terminal pro-BNP peptide (NT-pro-BNP). BNP is a hormone released by the myocardium in response to excessive stretching of cardiomyocytes, and NT-pro-BNP is its degradation derivative. BNP exhibits the biological activity, whereas no defined biological function has been found to be associated with NT-proBNP. The calculated biological half-lives of BNP range from 13 to 20 minutes and of NT-proBNP from 25 to 70 minutes. These molecules are widely used in clinical practice to ascertain presence of clinically suspected heart failure. Similarly to the other biomarkers mentioned so far, both BNP and NT-proBNP levels often vary according to comorbidities (i.e., atrial fibrillation, pericardial disease) and demographic characteristics like age, race and sex [54]. In 2002, Redfield *et al.* [55] observed that BNP levels were on average 32% higher in women with ejection fraction $>50\%$ if compared with their male counterparts. A more recent analysis confirmed these findings by evaluating levels of NT-pro-BNP, and additionally stratifying results for menopausal state of female participants [56]. In particular, men presented with lower NT-pro-BNP than women, and this relationship did not change after menopause, although pre-menopausal women administered with hormone replacement therapy did show higher levels of the biomarker if compared with those receiving no treatment. In both sexes, however, increasing NT-pro-BNP levels were related to decreasing free testosterone and increasing sex hormone binding protein concentrations. When accounting for these factors, all differences related to sex or hormone levels decreased considerably, suggesting that interaction between androgen levels and NT-pro BNP may explain the observed sex differences, a hypothesis formulated also in the Dallas Heart Study [57]. However, it should be noted that the most significant sex-related differences in terms of BNP levels were observed in the general population, whereas studies including acute patients reported only slight differences in the concentrations of this biomarker [58,59]. In general, women have been shown to present a tendency to lower increases of BNP and NT-pro-BNP in the acute setting when compared to their male counterparts. This phenomenon was confirmed in a meta-analysis by Mangusson *et al.* [60], which concluded that higher NT-pro-BNP levels were actually more strictly associated with incidence of heart failure in men than in women (hazard ratio [HR], 1.89; 95% CI, 1.75–2.05) in men versus 1.54; 95% CI, 1.37–1.74) in women [60]. It follows that further research is warranted to better define the clinical relevance of differences in BNP and NT-pro-BNP concentrations across sexes, and to establish whether the application of sex-specific thresholds would actually improve management and prognosis of patients in day-to-day practice.

Similar observations can be made on other biomarkers whose variations have been associated with an increased

risk of heart failure. For instance, galectin-3 is a pro-fibrotic protein that has been found to be significantly increased in patients with heart failure [61], and some levels of sex-related differences in its concentrations have been observed at baseline. Moreover, in a recent study conducted on data from 4 community-based cohorts with 12.5 years of follow-up, galectin-3 was significantly associated with incident heart failure only in women (HR: 1.13; 95% CI: 1.05–1.22), suggesting differences in the development of cardiac fibrosis as potential pathophysiological mechanisms underlying sex-related discrepancies in incidence and prognosis from heart failure [62]. More analyses still need to be conducted in order to definitely assess differences in its prognostic value between men and women [63].

6. Conclusions

With the rightful evolution of clinical and research perspective on women in cardiovascular diseases, female patients have gradually emerged as individuals, with pathophysiological characteristics and responses to treatment that differ profoundly to what has been observed in men and cannot simply be assumed on the basis of studies conducted on mostly or only male participants. The present review summarized the state-of-art knowledge on sex-related differences in cardiovascular risk factors and biomarkers expression of myocardial necrosis and dysfunction. What emerged is that while women do present significant differences either in indicators of inflammation, necrosis and ventricular dysfunction, the clinical and prognostic implications of these differences are less extensively characterized, a phenomenon that has prevented international organizations from adopting clear and standardized sex-specific thresholds for these indicators. Smoking and diabetes mellitus disproportionally increase the risk of obstructive CAD in women compared to men, and as so it may contribute to sex differences in outcomes from ACS. New studies conducted on contemporary, more inclusive cohorts are warranted to shed light on these issues and to bring us closer to a personalized and equal standard of care.

Author contributions

MB and OM conceived and designed the manuscript, drafted the manuscript, and revised it for important intellectual content. MS, RB and EC made substantial contributions to the conception and design of the manuscript, drafted the manuscript, and revised it for important intellectual content. All authors approved the final version of the manuscript.

Ethics approval and consent to participate

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Conflict of interest

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References

- [1] Gulati M. Yentl's Bikini: Sex Differences in STEMI. *Journal of the American Heart Association*. 2019; 8: e012873.
- [2] Ricci B, Cenko E, Vasiljevic Z, Stankovic G, Kedev S, Kalpak O, *et al*. Acute Coronary Syndrome: the Risk to Young Women. *Journal of the American Heart Association*. 2017; 6: e007519.
- [3] Cenko E, Ricci B, Kedev S, Vasiljevic Z, Dorobantu M, Gustiene O, *et al*. Reperfusion therapy for ST-elevation acute myocardial infarction in Eastern Europe: the ISACS-TC registry. *European Heart Journal. Quality of Care & Clinical Outcomes*. 2016; 2: 45–51.
- [4] Ricci B, Cenko E, Varotti E, Puddu PE, Manfrini O. Atypical Chest Pain in ACS: a Trap Especially for Women. *Current Pharmaceutical Design*. 2016; 22: 3877–3884.
- [5] Vaccarino V, Badimon L, Corti R, de Wit C, Dorobantu M, Manfrini O, *et al*. Presentation, management, and outcomes of ischaemic heart disease in women. *Nature Reviews. Cardiology*. 2013; 10: 508–518.
- [6] Healy B. The Yentl syndrome. *The New England Journal of Medicine*. 1991; 325: 274–276.
- [7] Bugiardini R, Cenko E. Sex differences in myocardial infarction deaths. *The Lancet*. 2020; 396: 72–73.
- [8] Shaw LJ, Bugiardini R, Merz CNB. Women and Ischemic Heart Disease. *Journal of the American College of Cardiology*. 2009; 54: 1561–1575.
- [9] Vaccarino V, Parsons L, Every NR, Barron HV, Krumholz HM. Sex-Based Differences in Early Mortality after Myocardial Infarction. *New England Journal of Medicine*. 1999; 341: 217–225.
- [10] Manfrini O, Cenko E, Bugiardini R. Gender Differences in Residual Risk Factors for Major Adverse Cardiovascular Events Following ACS and how to Bridge the Gap. *Current Atherosclerosis Reports*. 2020; 22: 65.
- [11] D'Onofrio G, Safdar B, Lichtman JH, Strait KM, Dreyer RP, Geda M, *et al*. Sex differences in reperfusion in young patients with ST-segment-elevation myocardial infarction: results from the VIRGO study. *Circulation*. 2015; 131: 1324–1332.
- [12] Bugiardini R, Ricci B, Cenko E, Vasiljevic Z, Kedev S, Davidovic G, *et al*. Delayed Care and Mortality among Women and Men with Myocardial Infarction. *Journal of the American Heart Association*. 2017; 6: e005968.
- [13] Khera S, Kolte D, Gupta T, Subramanian KS, Khanna N, Aronow WS, *et al*. Temporal Trends and Sex Differences in Revascularization and Outcomes of ST-Segment Elevation Myocardial Infarction in Younger Adults in the United States. *Journal of the American College of Cardiology*. 2015; 66: 1961–1972.
- [14] Cenko E, Yoon J, Kedev S, Stankovic G, Vasiljevic Z, Krljanac G, *et al*. Sex Differences in Outcomes after STEMI. *JAMA Internal Medicine*. 2018; 178: 632.
- [15] Nicholls SJ, Wolski K, Sipahi I, Schoenhagen P, Crowe T, Kapadia SR, *et al*. Rate of Progression of Coronary Atherosclerotic

- Plaque in Women. *Journal of the American College of Cardiology*. 2007; 49: 1546–1551.
- [16] Han SH, Bae JH, Holmes DR, Lennon RJ, Eeckhout E, Barsness GW, *et al.* Sex differences in atheroma burden and endothelial function in patients with early coronary atherosclerosis. *European Heart Journal*. 2008; 29: 1359–1369.
 - [17] Dart AM, Du X, Kingwell BA. Gender, sex hormones and autonomic nervous control of the cardiovascular system. *Cardiovascular Research*. 2002; 53: 678–687.
 - [18] Burke AP, Farb A, Malcom G, Virmani R. Effect of menopause on plaque morphologic characteristics in coronary atherosclerosis. *American Heart Journal*. 2001; 141: S58–S62.
 - [19] Padro T, Manfrini O, Bugiardini R, Canty J, Cenko E, De Luca G, *et al.* ESC Working Group on Coronary Pathophysiology and Microcirculation position paper on ‘coronary microvascular dysfunction in cardiovascular disease’ *Cardiovascular Research*. 2020; 116: 741–755.
 - [20] Cenko E, van der Schaar M, Yoon J, Manfrini O, Vasiljevic Z, Vavlukis M, *et al.* Sex-Related Differences in Heart Failure after ST-Segment Elevation Myocardial Infarction. *Journal of the American College of Cardiology*. 2019; 74: 2379–2389.
 - [21] Huxley RR, Woodward M. Cigarette smoking as a risk factor for coronary heart disease in women compared with men: a systematic review and meta-analysis of prospective cohort studies. *Lancet*. 2011; 378: 1297–1305.
 - [22] Yusuf S, Hawken S, Ounpuu S, Dans T, Avezum A, Lanas F, *et al.* Effect of potentially modifiable risk factors associated with myocardial infarction in 52 countries (the INTERHEART study): case-control study. *Lancet*. 2004; 364: 937–952.
 - [23] Manfrini O, Yoon J, van der Schaar M, Kedev S, Vavlukis M, Stankovic G, *et al.* Sex Differences in Modifiable Risk Factors and Severity of Coronary Artery Disease. *Journal of the American Heart Association*. 2020; 9: e017235.
 - [24] Dorobantu M, Onciul S, Tautu OF, Cenko E. Hypertension and Ischemic Heart Disease in Women. *Current Pharmaceutical Design*. 2016; 22: 3885–3892.
 - [25] Badimon L, Bugiardini R, Cenko E, Cubedo J, Dorobantu M, Duncker DJ, *et al.* Position paper of the European Society of Cardiology-working group of coronary pathophysiology and microcirculation: obesity and heart disease. *European Heart Journal*. 2017; 38: 1951–1958.
 - [26] Borgeraas H, Hertel JK, Svingen GFT, Seifert R, Pedersen EKR, Schartum-Hansen H, *et al.* Association of body mass index with risk of acute myocardial infarction and mortality in Norwegian male and female patients with suspected stable angina pectoris: a prospective cohort study. *BMC Cardiovascular Disorders*. 2014; 14: 68.
 - [27] Winzer EB, Woitek F, Linke A. Physical Activity in the Prevention and Treatment of Coronary Artery Disease. *Journal of the American Heart Association*. 2018; 7: e007725.
 - [28] Piepoli MF, Hoes AW, Agewall S, Albus C, Brotons C, Catapano AL, *et al.* 2016 European Guidelines on cardiovascular disease prevention in clinical practice. *European Heart Journal*. 2016; 37: 2315–2381.
 - [29] Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. *The New England Journal of Medicine*. 2000; 342: 836–843.
 - [30] Willerson JT, Ridker PM. Inflammation as a cardiovascular risk factor. *Circulation*. 2004; 109: I12–I10.
 - [31] Arnett DK, Blumenthal RS, Albert MA, Buroker AB, Goldberger ZD, Hahn EJ, *et al.* 2019 ACC/AHA Guideline on the Primary Prevention of Cardiovascular Disease. *Circulation*. 2019; 140: e596–e646.
 - [32] Khera A, McGuire DK, Murphy SA, Stanek HG, Das SR, Vongpatanasin W, *et al.* Race and gender differences in C-reactive protein levels. *Journal of the American College of Cardiology*. 2005; 46: 464–469.
 - [33] Rogowski O, Zeltser D, Shapira I, Burke M, Zakut V, Mardi T, *et al.* Gender difference in C-reactive protein concentrations in individuals with atherothrombotic risk factors and apparently healthy ones. *Biomarkers*. 2004; 9: 85–92.
 - [34] Clark DO, Unroe KT, Xu H, Keith NR, Callahan CM, Tu W. Sex and Race Differences in the Relationship between Obesity and C-Reactive Protein. *Ethnicity & Disease*. 2016; 26: 197–204.
 - [35] Lau ES, Paniagua SM, Guseh JS, Bhambhani V, Zanni MV, Courchesne P, *et al.* Sex Differences in Circulating Biomarkers of Cardiovascular Disease. *Journal of the American College of Cardiology*. 2019; 74: 1543–1553.
 - [36] Ford ES. C-reactive protein concentration and cardiovascular disease risk factors in children: findings from the National Health and Nutrition Examination Survey 1999–2000. *Circulation*. 2003; 108: 1053–1058.
 - [37] Shanahan L, Copeland WE, Worthman CM, Erkanli A, Angold A, Costello EJ. Sex-differentiated changes in C-reactive protein from ages 9 to 21: the contributions of BMI and physical/sexual maturation. *Psychoneuroendocrinology*. 2013; 38: 2209–2217.
 - [38] Lakoski SG, Cushman M, Criqui M, Rundek T, Blumenthal RS, D’Agostino RB, *et al.* Gender and C-reactive protein: data from the Multiethnic Study of Atherosclerosis (MESA) cohort. *American Heart Journal*. 2006; 152: 593–598.
 - [39] Rutter MK, Meigs JB, Sullivan LM, D’Agostino RB, Wilson PWF. C-reactive protein, the metabolic syndrome, and prediction of cardiovascular events in the Framingham Offspring Study. *Circulation*. 2005; 110: 380–385.
 - [40] Qasim AN, Budharaju V, Mehta NN, St Clair C, Farouk S, Braunstein S, *et al.* Gender differences in the association of C-reactive protein with coronary artery calcium in type-2 diabetes. *Clinical Endocrinology*. 2011; 74: 44–50.
 - [41] Cartier A, Côté M, Lemieux I, Périus L, Tremblay A, Bouchard C, *et al.* Sex differences in inflammatory markers: what is the contribution of visceral adiposity? *The American Journal of Clinical Nutrition*. 2009; 89: 1307–1314.
 - [42] Sobhani K, Nieves Castro DK, Fu Q, Gottlieb RA, Van Eyk JE, Noel Bairey Merz C. Sex differences in ischemic heart disease and heart failure biomarkers. *Biology of Sex Differences*. 2018; 9: 43.
 - [43] Hong GB, Gao PC, Chen YY, Xia Y, Ke XS, Shao XF, *et al.* High-Sensitivity C-Reactive Protein Leads to Increased Incident Metabolic Syndrome in Women but Not in Men: A Five-Year Follow-Up Study in a Chinese Population. *Diabetes, Metabolic Syndrome and Obesity*. 2020; 13: 581–590.
 - [44] Thygesen K, Alpert JS, Jaffe AS, Chaitman BR, Bax JJ, Morrow DA, *et al.* Fourth Universal Definition of Myocardial Infarction (2018). *Global Heart*. 2018; 13: 305–338.
 - [45] Eggers KM, Lindahl B. Impact of Sex on Cardiac Troponin Concentrations—a Critical Appraisal. *Clinical Chemistry*. 2017; 63: 1457–1464.
 - [46] Wiviott SD, Cannon CP, Morrow DA, Murphy SA, Gibson CM, McCabe CH, *et al.* Differential Expression of Cardiac Biomarkers by Gender in Patients with Unstable Angina/Non-ST-Elevation Myocardial Infarction. *Circulation*. 2004; 109: 580–586.
 - [47] Shah ASV, Griffiths M, Lee KK, McAllister DA, Hunter AL, Ferry AV, *et al.* High sensitivity cardiac troponin and the under-diagnosis of myocardial infarction in women: prospective cohort study. *British Medical Journal*. 2015; 350: g7873.
 - [48] Eggers KM, Johnston N, Lind L, Venge P, Lindahl B. Cardiac troponin i levels in an elderly population from the community—the implications of sex. *Clinical Biochemistry*. 2015; 48: 751–756.
 - [49] Gore MO, Seliger SL, Defilippi CR, Nambi V, Christenson RH,

- Hashim IA, *et al.* Age- and sex-dependent upper reference limits for the high-sensitivity cardiac troponin T assay. *Journal of the American College of Cardiology*. 2014; 63: 1441–1448.
- [50] Rubini Giménez M, Twerenbold R, Boeddinghaus J, Nestelberger T, Puelacher C, Hillinger P, *et al.* Clinical Effect of Sex-Specific Cutoff Values of High-Sensitivity Cardiac Troponin T in Suspected Myocardial Infarction. *JAMA Cardiology*. 2016; 1: 912–920.
- [51] Collinson PO, Saenger AK, Apple FS. High sensitivity, contemporary and point-of-care cardiac troponin assays: educational aids developed by the IFCC Committee on Clinical Application of Cardiac Bio-Markers. *Clinical Chemistry and Laboratory Medicine*. 2019; 57: 623–632.
- [52] Collet JP, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL, *et al.* 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation: The Task Force for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation of the European Society of Cardiology (ESC). *European Heart Journal*. 2021; 42: 1289–1367.
- [53] Vargas KG, Tajsic M, Latsuzbaia A, Andric T, Kassem M, Jaeger B, *et al.* Sex-based differences of copeptin for early rule-out of non-ST-elevation myocardial infarction. *European Heart Journal*. 2020; 41.
- [54] Bozkurt B, Coats A, Tsutsui H. Universal Definition and Classification of Heart Failure. *Journal of Cardiac Failure*. 2021; S1071-9164: 00050-6.
- [55] Redfield MM, Rodeheffer RJ, Jacobsen SJ, Mahoney DW, Bailey KR, Burnett JC. Plasma brain natriuretic peptide concentration: impact of age and gender. *Journal of the American College of Cardiology*. 2002; 40: 976–982.
- [56] Lam CSP, Cheng S, Choong K, Larson MG, Murabito JM, Newton-Cheh C, *et al.* Influence of sex and hormone status on circulating natriuretic peptides. *Journal of the American College of Cardiology*. 2011; 58: 618–626.
- [57] Chang AY, Abdullah SM, Jain T, Stanek HG, Das SR, McGuire DK, *et al.* Associations among androgens, estrogens, and natriuretic peptides in young women: observations from the Dallas Heart Study. *Journal of the American College of Cardiology*. 2007; 49: 109–116.
- [58] Franke J, Lindmark A, Hochadel M, Zugck C, Koerner E, Kepler J, *et al.* Gender aspects in clinical presentation and prognostication of chronic heart failure according to NT-proBNP and the Heart Failure Survival Score. *Clinical Research in Cardiology*. 2015; 104: 334–341.
- [59] Dewan P, Rørth R, Jhund PS, Shen L, Raparelli V, Petrie MC, *et al.* Differential Impact of Heart Failure with Reduced Ejection Fraction on Men and Women. *Journal of the American College of Cardiology*. 2019; 73: 29–40.
- [60] Magnussen C, Niiranen TJ, Ojeda FM, Gianfagna F, Blankenberg S, Vartiainen E, *et al.* Sex-Specific Epidemiology of Heart Failure Risk and Mortality in Europe. *JACC: Heart Failure*. 2019; 7: 204–213.
- [61] Ho JE, Liu C, Lyass A, Courchesne P, Pencina MJ, Vasan RS, *et al.* Galectin-3, a marker of cardiac fibrosis, predicts incident heart failure in the community. *Journal of the American College of Cardiology*. 2012; 60: 1249–1256.
- [62] Suthahar N, Lau ES, Blaha MJ, Paniagua SM, Larson MG, Psaty BM, *et al.* Sex-Specific Associations of Cardiovascular Risk Factors and Biomarkers with Incident Heart Failure. *Journal of the American College of Cardiology*. 2020; 76: 1455–1465.
- [63] Suthahar N, Meems LMG, Ho JE, Boer RA. Sex-related differences in contemporary biomarkers for heart failure: a review. *European Journal of Heart Failure*. 2020; 22: 775–788.