

## Extended abstract from IGM webinar May 6, 2021 Gender differences in COVID-19 pandemic around the world

### Metabolic issues during the COVID-19 pandemic: gender difference

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The coronavirus disease 2019 (COVID-19) has rapidly spread all over the world, causing a great number of casualties. From the very beginning of the pandemic, it has become apparent that there are multiple risk factors associated with an increased risk of disease severity and death. These include older age, smoking and several underlying comorbidities, as well as gender.<sup>1</sup>

Susceptibility for SARS-CoV-2 infection appears to be similar in men and women, and yet most of the clinical and epidemiological data has shown that almost twice as many men with COVID-19 suffer severe symptoms or death as women.<sup>2</sup> Despite a similar incidence between the two genders, men consistently show a more severe phenotype and an increased mortality rate (62.4%) across age groups at global level.<sup>3</sup> A large population-based study performed in England, which included over 17 million adults and 10,926 COVID-19-related deaths, found that males had a significantly higher risk of death (HR 1.59; 95% CI, 1.53-1.65) than females.<sup>4</sup> A recently published review reported that, overall, males account for 59-75% of all COVID-19 deaths.<sup>5</sup>

Sexual dimorphism in COVID-19 should not come as a surprise, because it is well known that men and women respond to viral infections differently, as already reported during other flu outbreaks.<sup>6</sup> Many of the genes playing a key role in the immune response are located on the X chromosome, including those involved in determining the innate and adaptive immune responses to viral infections.<sup>7</sup> Interestingly, gene encoding for the ACE2 receptor – through which SARS-CoV-2 binds to the cell membrane and enters the host cell – is also located on the X chromosome, so that a higher degree of protein expression could be expected in the female gender, which may increase the risk of viral infection.<sup>8</sup> However, a higher ACE2 activity – particularly in the lungs and in the cardiovascular system – has been claimed to confer some protection, which may account for the less severe form of COVID-19 in women.<sup>9</sup> Consistent with this hypothesis is the finding that the male heart has less ACE2-expressing cells than the female one,<sup>10</sup> which provides support to a sex-specific regulation of ACE2. Nevertheless, such sex-dependent ACE2 expression has not yet been validated in humans, and

no relevant influence of medications such as ACE-inhibitors has been documented.

Sex differences in the manifestation of infectious diseases have long been attributed also to the influence of sex hormones. Experimental work performed in a murine model of SARS-CoV-2 infection has shown that male animals were more susceptible to infection and had higher mortality than females. Interestingly, the estrogen deprivation obtained by ovariectomy nullified this protection, causing an increase in mortality.<sup>11</sup> These results indicate how the balance between androgens and estrogens is likely to play an important role in modulating immune responses in coronavirus infections. Conversely, men receiving androgen deprivation therapy seem to be protected from SARS-CoV-2 infection, which further supports the concept of sexual dimorphism in response to the SARS-CoV-2 infection.<sup>12</sup> A gene expression study on the immune system of mice indicated that this sexual dimorphism is mainly limited to macrophages, with an up-regulation of macrophage-specific genes (eg., complement-related and IFN-stimulated genes) found in female cells.<sup>13</sup> Thus, females could show a more activated innate response pathway prior to an infection with a pathogen. Furthermore, TLR7 signaling and IFN production seem to be more expressed in females, while estrogen also increases TLR7 expression. Finally, the immune system has been implicated in driving a detrimental and dysregulated inflammation in COVID-19,<sup>14</sup> therefore it may seem counterintuitive that males are at greater risk of COVID-19 hyperinflammation, considering that females have been described to mount stronger immune responses to viral infections. This highlights once again the complexity of the differences between the male and female immune systems, and their responses to infection.

In summary, infection rates appear to be similar between men and women, although the response to infection differs between the sexes. It has been suggested that anti-viral responses and viral clearance, mediated by IFN and TLR7, are increased in females, contributing to the reduced COVID-19 mortality observed in women compared with men. In men, dysregulated inflammation and an increased cytokine release are likely to be responsible for the increase in ARDS, respiratory failure

and cardiovascular comorbidities, as well as a higher mortality.

Together with male gender, also diabetes – and the degree of glycemic control – have been shown to be associated with a poorer prognosis and higher mortality in COVID-19 patients.<sup>15</sup> According to the analysis of Scully and colleagues,<sup>16</sup> the prevalence of T1DM (1.8 vs 1.8%) and T2DM (38 vs 36%) did not differ between males and females with SARS-CoV-2 infection. Similarly, de Jong et al. reported a comparable association between diabetes and COVID-19 death in both sexes,<sup>17</sup> while a study carried out in England in 61 million subjects showed a higher mortality in diabetic women than in men.<sup>18</sup>

To gain more insights about the potential differences in the inflammatory and metabolic parameters between male and female diabetics, we analyzed a cohort of 271 subjects admitted to our hospital because of COVID-19 and in whom we reported a higher mortality amongst those with hyperglycemia – without known diabetes – upon admission.<sup>19</sup> In this cohort, the distribution of subjects with normo- or hyperglycemia upon admission, or with known diabetes, was similar between the two genders (males: 51, 27 and 22%; females: 62, 19, 19%, respectively). Among the subjects with normoglycemia (93 males and 56 females), women were older, and had higher levels of total and LDL cholesterol, but lower levels of ferritin and pro-calcitonin. In the subjects with hyperglycemia upon admission, without known diabetes, the only difference was once again a higher level of cholesterol. Finally, diabetic women had lower levels of ferritin and higher levels of PaO<sub>2</sub>/FIO<sub>2</sub>. In males only, hyperglycemia upon admission was associated with a reduced in-hospital survival, while in women both hyperglycemia and diabetes upon admission were associated with a higher mortality. These results should be interpreted with caution, due to the small number of individuals with diabetes in our cohort (70 males and 30 females). To extend this analysis to a larger number of subjects, Thakur et al. compared the features and outcomes of multiple studies, distinguishing between preponderant male and preponderant female studies.<sup>20</sup> According to their analysis, the pooled proportion of subjects with diabetes and COVID-19 was similar in male and female preponderant studies with respect to presence of comorbidities (0.19 vs 0.16;  $p = 0.112$ ), severity of disease (0.41 vs 0.48;  $p = 0.414$ ) and mortality (0.35 vs 0.29,  $p = 0.189$ ).

A final aspect to be considered concerns the potential effects of glucose-lowering agents on the COVID-19 outcomes. This was initially suggested by the report of Bramante et al., which showed that women – but not men – receiving metformin at the time of their hospitalization had lower COVID-19 mortality (HR 0.78, 95% CI 0.65-0.95).<sup>21</sup> This finding was interpreted as a confirma-

tion of the experimental data obtained both from animals and humans, showing a sexual dimorphism of metformin with respect to its anti-inflammatory effect.<sup>21</sup> However, the results of this study, as well as other similar ones, should be taken with great caution, due to the retrospective nature of the analyses, the partial information available with regard to the duration of treatments, and the dose exposure of glucose-lowering agent, as well as the potential confounding by indication. Similarly, the results of a recent large analysis providing evidence of an association between the prescription of some glucose-lowering drugs and COVID-19-related mortality should also be interpreted in light of these considerations.<sup>22</sup> Interestingly, however, no difference could be found between male and female subjects with respect to the potentially positive or negative effects of the glucose-lowering agents considered.

In conclusion, although the current evidence does not suggest a gender prevalence for SARS-CoV-2 infection, males show more severe forms of COVID-19 and a greater mortality rate. These differences do not seem to be accounted for by the different expression of ACE2, although a sexual dimorphism has been postulated, and the genetic regulation and hormonal influence on the immune response in males and females are likely to confer some degree of protection to the latter. Diabetes, however, seems to overrule such sexual dimorphism, although the exact underlying mechanisms still has to be clarified.

**Keywords.** COVID-19, SARS-CoV-2, gender, sex-difference, diabetes.

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