Documenting Sex and Sex Differences in Animal Studies

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The National Institutes of Health reports that women now represent nearly half of all participants in clinical research (https://grants.nih.gov/grants/guide/notice-files/NOT-OD-15-102.html). At *Thrombosis and Haemostasis*, we have indeed published several clinical articles, specifically highlighting the effects of gender differences in recent years.[1–12] In contrast, basic and preclinical studies are more often and at times traditionally conducted in male animals. This can lead to an underappreciation of sex differences and their influence in health and disease, despite sex being not only an important experimental variable but also a notable factor determining cardiovascular outcomes, for example, stroke risk in atrial fibrillation, as well as platelet function and antiplatelet therapy in women.[3,4,6,11–14] Hence, we would like to follow the good example implemented by the Arteriosclerosis, Thrombosis, and Vascular Biology Council[15,16] and commit ourselves to regularly monitoring sex and sex differences in animal studies published in *Thrombosis and Haemostasis*. This report should help elaborating an editorial approach regarding the requirements for documenting sex when conducting animal studies to be submitted to our journal.

In our first survey, we counted 59 articles reporting animal studies in *Thrombosis and Haemostasis* spanning 2018 and 2019.[17–75] All studies, except two, were conducted in mice or rats. The two other studies used either rabbits or nonhuman primates. Mice represented the vast majority (86%) of animals used. One article studied mouse embryos and was not included. Out of the remaining 58 articles, 52% reported sex consistently, 38% did not report sex at all, while 10% reported sex albeit not for all animals used (►**Fig. 1**). Among the studies documenting sex, 53% used male animals only, 14% of studies reported females only, whereas 33% included both male and female animals, albeit not necessarily for all experiments (►**Fig. 2**). None of the articles studying one sex only provided a justification for it. Out of the articles investigating both sexes, less than half of the studies ($N = 5$) set out to compare results in both sexes. The other studies either pooled data from both sexes or used one or another sex depending on the experiment without providing justification for this. One study only provided justification for pooling data of both sexes together.

This first analysis clearly warrants adjusting our editorial process to improve documentation and justifications of sex and sex differences in animal studies published in the journal as an incentive to improve the potential mechanistic and/or clinical relevance of the results. As a consequence, we have decided that future publications will require systematic documentation of the sex used in preclinical studies with immediate effect. We should strive to control that this consistently applies to all animals used in each study. We indeed noted that 10% of studies reported sex for one mouse background or one species but not for other backgrounds/species within the same study. We should also demand and control convincing explanation for studying only one sex, from either the scientific literature, preliminary data, or other relevant considerations. None of the articles analyzed here provided such a justification. Thus, the Editors should encourage preclinical studies to include both sexes, and presenting data distinctly for each sex to potentially identify sex differences wherever possible. Pooled data was only briefly justified in one article analyzed

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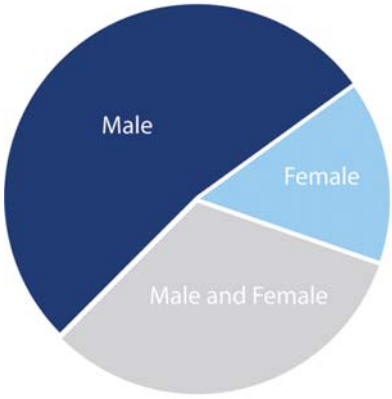
Sex reported consistently N (%)	Sex reported inconsistently N (%)	Sex not reported N (%)
30 (52%)	6 (10%)	22 (38%)

Fig. 1 Reporting sex in animal studies published in *Thrombosis and Haemostasis* in 2018–2019. The Pie Chart shows the percentage of manuscripts not reporting sex (grey slice), reporting sex in animal studies consistently (dark blue slice) and inconsistently (light blue slice).



Male N (%)	Female N (%)	Male and female N (%)
19 (53%)	5 (14%)	12 (33%)

Fig. 2 Sex used for animal studies published in Thrombosis and Haemostasis in 2018–2019 when documented. The Pie Chart shows the percentage of manuscripts using male and female (grey slice), male only (dark blue slice) and female only (light blue slice).

here. Pooling data of both sexes together should only be permitted when results do not reveal significant differences between male and female animals.

In conclusion, we will accordingly modify our submission guidelines and request authors to report sex and study both sexes whenever possible to ensure that this important aspect is no longer ignored. We are not only confident this should improve transparency and reproducibility of data published in our journal, but expect this new incentive to provide important cues to understand sex influences on cardiovascular homeostasis and disease.

Conflict of Interest

G.Y.H.L. reports consultancy and speaker fees from Bayer, Bayer/Janssen, BMS/Pfizer, Biotronik, Medtronic, Boehringer Ingelheim, Microlife, Roche, and Daiichi-Sankyo outside the submitted work. No fees received personally.

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