

Complete checklist for the study based on STROBE:

Title and abstract:

1. The title accurately reflects the content and scope of the study. The abstract provides a clear summary of the background, methods, results, and conclusions of the study.

Example: The abstract includes information on the prevalence of AAS use, its association with adverse health effects, the rationale for the study, the study design, and key findings related to cardiovascular and mental health outcomes.

Introduction:

2. Background/Rationale: The introduction provides a concise overview of the background information and rationale for the study, including relevant literature.

Example: The introduction discusses the prevalence of AAS use, its association with cardiovascular and mental health outcomes, and the gaps in existing research regarding the effects of AAS on different health parameters.

3. Objectives/Hypotheses: The specific objectives or hypotheses of the study are clearly stated.

Example: The study aims to investigate the associations between AAS use and various health outcomes, including preclinical cardiovascular disease, circulating risk markers, and mental health problems, among recreational athletes.

Methods:

4. Study Design: The study design (e.g., nationwide cross-sectional cohort study) is clearly described.

Example: The study describes a nationwide cross-sectional cohort study conducted in Denmark to investigate the associations between AAS use and different health outcomes.

5. Setting: The setting where the study took place is clearly described.

Example: The study is conducted at Odense University Hospital, Denmark, with participants recruited from recreational athletes engaged in different forms of physical activity.

6. Participants: Details about the study participants, including eligibility criteria, recruitment methods, and sample size determination, are provided.

Example: Participants include current and former AAS users as well as physically active control subjects matched for sex and age, recruited through targeted announcements, social media posts, and other recruitment strategies.

7. Variables: All variables investigated in the study, including exposure, outcomes, and potential confounders, are clearly defined.

Example: Variables include AAS use patterns, cardiovascular outcomes (e.g., carotid and femoral plaque development, myocardial function), mental health parameters, and demographic characteristics.

8. Data Sources/Measurement: The sources of data and methods of measurement for each variable are described.

Example: Data sources include interviews, questionnaires, physical examinations, laboratory tests (e.g., urine and blood samples), and imaging studies (e.g., echocardiography, ultrasound).

9. Bias: Potential sources of bias and strategies to address them are discussed.

Example: The study discusses potential biases related to participant recruitment, measurement techniques, and confounding variables, with strategies to minimize bias.

10. Study Size: The rationale for the sample size determination is provided, and the actual sample size is reported.

Example: The study aims to include at least 120 participants in the pilot study, with recruitment efforts ongoing for the larger cohort study.

11. Quantitative Variables: Methods for handling quantitative variables (e.g., means, standard deviations) are described.

Example: Descriptive statistics will be used to summarize demographic and clinical characteristics, with correlation analyses and regression models to explore associations between AAS use and health outcomes.

12. Statistical Methods: The statistical methods used to analyze the data are clearly described, including any adjustments for confounding variables.

Example: Statistical methods include one-way ANOVA, Chi-squared tests, Kruskal Wallis test, correlation analyses, and multivariable regression models to assess associations and adjust for potential confounders.

Other Information:

13. Funding: Sources of funding for the study are disclosed.

Example: The study acknowledges funding support from relevant institutions or organizations, along with any potential conflicts of interest among the study authors.

14. Ethics Approval: Ethical approval for the study is acknowledged.

Example: The study acknowledges approval from the Regional Committee on Health Research Ethics for Southern Denmark and compliance with ethical standards outlined in the Declaration of Helsinki.

15. Conflict of Interest: Any potential conflicts of interest among the study authors are disclosed.

Example: The study discloses any financial or personal conflicts of interest that may influence the research or interpretation of findings.

16. Availability of Data and Materials: Information on the availability of data and materials related to the study is provided.

Example: The study outlines plans for data sharing and availability of materials for future research or replication efforts.

17. Acknowledgments: Any individuals or organizations that contributed to the study but do not meet authorship criteria are acknowledged.
Example: The study acknowledges contributions from participants, research assistants, funding agencies, and other individuals or organizations that supported the research endeavor.

Study outcomes

| Investigations | Pilot study [a] | Subsequent cohort study [b] |
|---|-----------------|-----------------------------|
| Non-contrast coronary CT | X | (X) |
| Coronary CT angiography | X | (X) |
| Echocardiography | X | (X) |
| Ultrasound of the carotid and femoral arteries | X | (X) |
| Retinal fundus photo | X | (X) |
| Grip strength measurement | X | (X) |
| Bioelectrical impedance | X | X |
| Blood samples - immediate analyses + later analyses | X | X |
| Urine samples | X | X |
| Clinical examination | X | X |
| Medical history and socioeconomic status | X | X |
| Questionnaires | X | X |