diminished, compared to the 10% KSR group.
Conclusion: In summary, the combination of 5% KSR and 5% PRGF failed to support complete in vitro spermatogenesis and instead promoted cellular degeneration and apoptosis. The reduced expression of key germ cell and proliferation markers, alongside increased apoptotic signaling, suggests that this medium disrupts the microenvironment required for germ cell development. These results suggest that the KSR and PRGF combination, in the tested ratio, is not an appropriate formulation for maintaining or promoting spermatogenic progression *in vitro*.
Keywords: *In Vitro* Spermatogenesis, PRGF, KSR, Spermatogonial Stem Cells, Organ Culture

P-87: Systematic Review of Animal Models in Reproductive Medicine: Opportunities and Challenges in Studying Human Infertility

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Background: Animal models constitute an essential component for the advancement of reproductive medicine. They provide a necessary framework to study the molecular and physiological mechanisms of human infertility. Given the complexity of the human reproductive systems and ethical restrictions on experimentation in humans, the animal models thus become useful for simulating different pathological and physiological reproductive states. These models help to understand disease etiology and play an invaluable role in developing novel therapeutic interventional strategies.
Materials and Methods: This study is systematic review. Three scientific databases, namely PubMed, Scopus and Web of Science, were used for an extensive search for placement. A total of 5194 articles have been found at the beginning. After eliminating duplicate and irrelevant studies, titles and abstracts were screened independently by two reviewers. Selected full-text articles were proof-read according to the previously spelt inclusion and exclusion criteria. In the end, all disagreements between reviewers were solved through discussion.
Results: Ovarian physiology, spermatogenesis, embryogenesis, as well as the efficacies of assisted reproductive technologies (ARTs) have been studied using a variety of animal models: rodents (mice and rats), non-human primates, and domestic species such as sheep and pigs. Each of these models has its own advantages regarding the mimicry of human reproductive biology. Inter-species variations, ethical issues, and limited translational possibilities remain critical impediments. Yet, in the face of these obstacles, there are a number of models with relatively evident potential for pre-clinical research.
Conclusion: The strategic selection and rigorous standardization of animal models can significantly increase their relevance and reliability in human reproductive research. No model can truly reproduce human physiology, but these systems remain one of the viable ways in understanding infertility and ameliorating ART outcomes. Genetic engineering and cross-species modeling may provide potential solutions to present shortcomings in the foreseeable future.
Keywords: Animal Models, Reproductive Biology, Human Infertility, Assisted Reproductive technologies, Translational Research

P-88: Application of Beetroot-Derived Natural Dye as An Alternative to Eosin-Nigrosin for Human Sperm Morphology Assessment

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Background: Background: Natural dyes are increasingly being used in biomedical applications because of their low toxicity, biodegradability, and sustainability compared to synthetic dyes. Traditional stains, such as eosin-nigrosin, used for sperm evaluation, raise concerns about chemical toxicity. This study evaluated the efficacy of a beetroot (Beta vulgaris)-derived natural dye as a non-toxic alternative dye for human sperm morphology assessment.
Materials and Methods: Beetroot pigment was extracted by homogenizing fresh beetroot in 70% ethanol (1:5 w/v), followed by agitation for 24 h at 4°C. After centrifugation (3000 rpm, 15 min) and filtration (Whatman No.1), the extract was diluted to 20% (v/v) with normal saline for staining. Ten air-dried human sperm smears (from two donors; all cells were non-viable) were stained with beetroot solution for 15 min at room temperature, then rinsed with PBS (pH=7.4) without fixation. Sperm morphology was exclusively evaluated using the WHO criteria by brightfield microscopy (1000× magnification). Eosin-nigrosin-stained slides were used for comparison.
Results: The beetroot dye successfully stained the sperm head, midpiece, and tail, providing distinct visualization and color contrast adequate for morphological differentiation according to the WHO guidelines. Importantly, the dye maintained structural integrity without causing head swelling or significant artifacts. Visual clarity for morphological assessment was comparable to that of traditional eosin-nigrosin staining. The staining method exhibited good stability.
Conclusion: Beetroot extract has significant potential as a natural, safe, and sustainable histological stain for human sperm morphology analysis. This offers an appropriate method for routine laboratory assessments. Further validation may support its integration into clinical practice.
Keywords: Beetroot Dye, Sperm Morphology, Natural Stain, Eosin-Nigrosin

P-89: The Effect of Human Follicular Fluid on Indices of Sperm and Oxidative Stress in Normospermic Men during Cryopreservation Process

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Background: Today, sperm cryopreservation is one of the most common approaches in assisted reproductive techniques (ART)