

Integrated Method for Mouse Identification and Genotyping: An Innovative Approach Combining Animal Welfare, Scientific Reliability, and Regulatory Compliance

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Context

Individual identification = key for monitoring, reproducibility, and ethics

Current mouse identification techniques:

- 🐭 **Electronic chips** (implanted under the skin of mice)
- 🐭 **Tattoos** (usually ink-based, applied to the skin)
- 🐭 **Ear notching / punching** (perforation of mouse ears)
- 🐭 **Toe clipping** (used in young mice)

⚠️ Associated issues:

- 🐭 **Animal stress:** Direct impact on animal well-being and potential alteration of experimental results.
- 🐭 **Inflammation risks:** Inflammation risks: Procedures may induce local or systemic inflammatory responses, affecting the mice.
- 🐭 **Invasive methods:** Some techniques require manipulations that can cause pain, discomfort, or post-operative complications.
- 🐭 **Regulatory challenges:** Need to strictly comply with ethical protocols and applicable legislation (e.g., European Directive 2010/63/EU), with increasing demands for justification and reduction of harm.

Genotyping= A Scientific Imperative

Objective: Verify the genetic lineage of mice via PCR.

🐭 **Tail (biopsy)**

Sample of 2 to 5 mm. Provides a lot of DNA. Used from 10–14 days.

🐭 **Ear (perforation)**

Tissue collected during identification. Less invasive. DNA can sometimes be limited.

🐭 **Hair**

Non-invasive method. Easy, but DNA can sometimes be weak.

🐭 **Buccal/Rectal Swabs**

Minimally invasive. Well tolerated. DNA varies depending on extraction.

⚠️ Associated Issues:

- 🐭 **Variable level of invasiveness**
- 🐭 **Impact on sample quality & animal welfare**
- 🐭 **Regulatory status sometimes restrictive (e.g., tail cutting)**

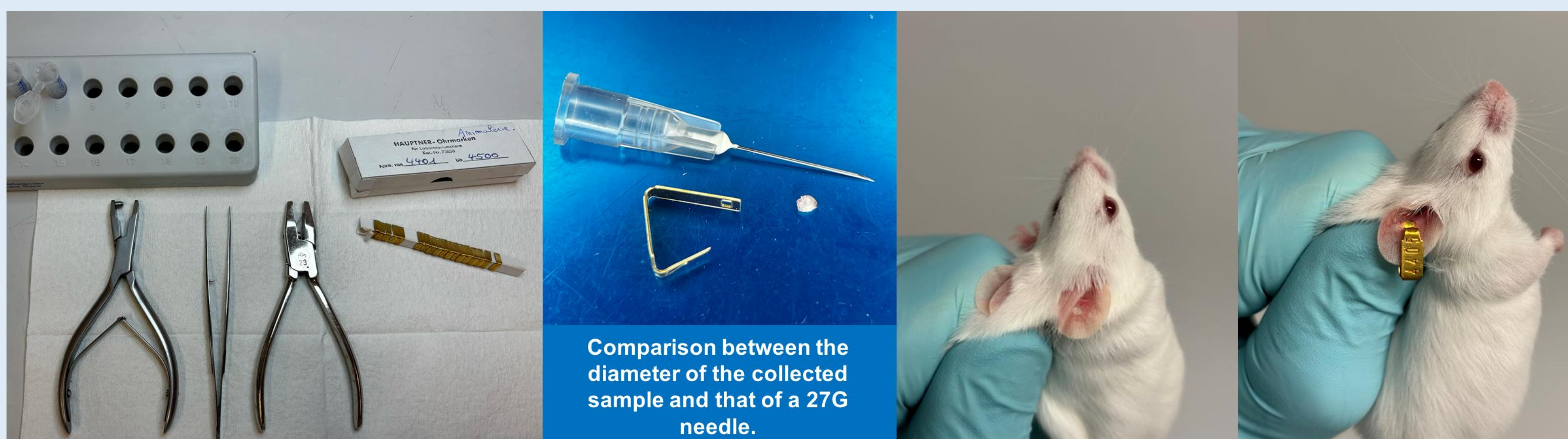
Comparative Summary

Method	Invasiveness	Recommended Age	DNA Quality	Ease of Use	Ethical Considerations	Notes
Tail Snip	Invasive	Newborn to adult	Good quality (high yield)	Moderate (requires expertise)	Ethical concerns due to invasiveness and potential pain	Widely used for genotyping, provides high-quality DNA.
Toe Clipping	Invasive	Newborn to juvenile	Good quality (high yield)	Easy to perform	High ethical concerns; can cause pain and affect mobility	Less commonly used today due to ethical concerns. Historically used in genetic studies.
Buccal Swab	Non-invasive	Neonates to adults	Moderate quality (lower yield)	Very easy and quick	Low ethical concerns, causes minimal stress to animals	Suitable for large-scale studies; DNA quality is lower compared to other methods.
Ear Punch/Notch	Non-invasive	Neonates to adults	Good quality (high yield)	Easy to perform	Minimal ethical concerns	Primarily used for identification in colony management.
Blood Sampling	Invasive (mild)	Juvenile to adult	Excellent quality (high yield)	Moderate (requires expertise)	Moderate ethical concerns due to blood loss and stress	Ideal for detailed genetic analysis; requires careful handling to minimize animal stress.



Our Integrated Approach: A One-Step Solution

- 🐭 **Ear Punching** (1 to 2 mm) → Sample for PCR.
- 🐭 **Immediate placement** of an identification loop in the created hole.



Major Advantages

Aspect	Advantages
🐭 Animal welfare	Less handling → reduced stress
🌿 Prevention	Fewer inflammatory reactions
🧠 Psychological effects	Fewer repeated restraints
🔬 Scientific quality	Reliable samples for PCR
📋 Regulatory compliance	Method not classified as an experimental procedure
📊 Impact on statistics	Proposed alternative for genotyping non-experimental mice, currently performed by tail biopsy.

Conclusion

"One procedure, two objectives: animal welfare and scientific rigor."

The integration of identification and genotyping:

- 🐭 **Reduces animal stress** by optimizing procedures and minimizing disruptions.
- 🐭 **Improves the quality of scientific data** through precise identification and better sample management.
- 🐭 **Fully aligns with ethical and legal recommendations**, ensuring compliance with animal welfare standards while maintaining the necessary scientific rigor.

Sample Collection for Genotyping and Ear Loop Placement

Restraint of the Mouse

- 🐭 Gently but firmly restrain the mouse to minimize stress and ensure safety.
- 🐭 Use an appropriate and humane grip (e.g., by the nape of the neck) to limit excessive movement, while maintaining the mouse's comfort and well-being.
- 🐭 **Ear Sample Collection for Genotyping**
- 🐭 Begin by disinfecting the sampling area to prevent contamination and infection.
- 🐭 Use a sterile micro-punch to obtain a small tissue sample from the ear, ensuring precision.
- 🐭 Perform the procedure carefully and gently to minimize discomfort, taking care to avoid any injury to the animal.

Ear Loop Placement

- 🐭 Prior to inserting the ear loop, carefully prepare the perforated area to ensure a smooth insertion.
- 🐭 Gently insert the loop, ensuring that it is securely attached, while ensuring it does not interfere with the mouse's natural movement.
- 🐭 Verify that the loop is properly fixed in place and does not cause any discomfort or restrict the mouse's activity.

Criteria	Ear punch for genotyping and identification looping
Invasiveness	Less invasive, does not alter the individual's physiology
Pain for the animal	Mild (faster and less painful)
Risk of infection	None if proper disinfection
Cost and speed	Quick and convenient, requires specialized equipment (inexpensive)
Repeatability	Can be repeated without compromising the animal
Long-term impact	Limited short-term discomfort, no impact on overall condition.