



APPROVED COLLECTION METHOD OVERVIEW

Approved Collection Protocols Requirements

Orivet

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Ensuring sample integrity is a key part of any testing and more importantly eliminates any doubt on the test result being from that animal. Many organisations allow individuals to collect and send samples in themselves. Orivet allows for samples to be submitted directly by the owner, a veterinarian or an approved collection agent. Orivet reports results as, **Approved Collection Method (ACM) – Yes or No** depending on who has collected the sample. If the animal submitted has been collected independently (not by the owner) and identified via its microchip number, then the report will identify this as being Approved Collection Method – **Yes**.

Positive Identification and **Independent Collection** are seen as two key protocols to ensure sample integrity and internationally accepted results without any doubt on the sample submitted. These two requirements are discussed below.

1. POSITIVE IDENTIFICATION

Positive Identification (Positive ID) is the identification of a dog via its **microchip number** or a **recognised tattoo scheme** at the time of sample collection. This is a key component of our programme as it ties all results to the unique microchip number for the dog presented for DNA sample collection and examination.

All Approved Collection Method results must have the sample submitted identified via Positive ID. Positive ID **MUST** be cited using a microchip scanner to locate the microchip and record the number. The collector must acknowledge the number, record it and is required to sign off on the application form that they have identified the animal via Positive ID.



In cases where the owner presents documentation eg. Pedigree or in cases where a microchip cannot be scanned the sample collector must note that the “microchip could not be found/scanned” on the space left for the microchip number and submit the sample. A copy of the registration paper should be accompanied with the sample.

Orivet Recommendation: Where possible the owner should present and show a copy of the registration (pedigree) paper at the time of collection, although not a requirement it assists in confirming the dog’s microchip and registration details.

2. INDEPENDENT COLLECTION

Independent collection is also a requirement of an ACM genetic results. Samples must be collected independently for any such results to be issued. All veterinarians are automatically accepted and approved as independent collection officers. They require no further accreditation.

To assist breeders with sample collection, Approved Collection Agents will be part of the programme and accepted as independent collectors. It is proposed that these Agents will be nominated by member bodies or breed clubs and be part of an accredited/training scheme. ORIVET GENETIC PET CARE will encourage individuals who have a basic understanding of genetic screening for their breed and more importantly have shown genetic testing to be part of their breeding programme. Training will focus on ensuring all non-vets attend a course to be developed by ORIVET GENETIC PET CARE; these will be run locally at ORIVET GENETIC PET CARE and through webinars. All Collection agents MUST attend an annual webinar on sourcing relevant genetic information, basic genetics and a demonstration on how to carry out a collection.



An exam is also sent to all webinar participants and this must be completed and submitted as part of the accreditation process. Once completed participants are issued with a Collection Agent ID number. This number is used as the main identifier and must be cited when carrying out a collection. This number is used as a means of approved collection identification and reporting any collection which do not follow approved collection processes. Collectors details appear in the DNA report in the section titled – Collection Details. All collection agents are listed on the Orivet web site.

Please Note: Collection Agents **are not** permitted to collect from any of their own dogs, dogs that they co-own, dogs that they will use for breeding (stud or bitch), or dogs from any family members. If any such samples are collected and submitted that WILL NOT meet the requirements of an approved collection.

Collection Kits are made freely available to all collection officers and can also be ordered electronically though the ORIVET GENETIC PET CARE website. All collection officers MUST have access to a microchip reader for collections, ORIVET GENETIC PET CARE will have a small supply for loan to carry out collections. Collection Agents will receive regular updates via email including new disease releases, fact sheets and price changes. All Collection procedures will be made available electronically.

Collection instructions are included with all collection kits as part of the DNA Collection Kit.



Below is a summary of the various sample submission possibilities and the approved collection outcomes.

MICROCHIP NO CITED	COLLECTED BY	APPROVED COLLECTION METHOD
YES	OWNER	NO ❌
NO	OWNER	NO ❌
NO	VET/APPROVED COLLECTOR	NO ❌
YES	VET/APPROVED COLLECTOR	YES ¹ ✅

¹ Completed ACM form must accompany sample

Approved Method Reconfirmation

The requirement for positive identification does not impose any additional costs to the breeder as it is compulsory in many states to microchip all animals prior to sale. Owners do have the option to have their puppy screened prior to microchipping to allow them to select puppies based on known genetic disease status or to confirm parentage prior to registration purposes. These results are reported as ACM – No. Orivet allows an owner to have these re-submitted for DNA Profile and an Approved Collection Method after microchipping. A new sample **MUST** be collected and submitted and Orivet DNA profile the new sample to verify it is identical. This is done at a reduced fee to the owner. Sample is submitted as a “approved collection method - reconfirmation”. The sample is run and the DNA Profile analysed to identify if it is the same animal submitted without a microchip number.

Please Note: Genetic testing is still accepted and can be carried out without any form of positive identification results will be reported as Approved Collection Method - No.



3. INDEPENDENT COLLECTION

Samples accepted for genetic testing include buccal (cheek) swabs, semen and blood.

3.1 Orivet collection kits consist of sterile nylon brushes – pack of 3 – with 4 unique barcodes (6 digits). Buccal swabs are the preferred method of sample collection for genetic testing. Sterile cotton swabs are also accepted but incur an extra charge – handling fee. Orivet collection kits are supplied free of charge upon request.

COLLECT DNA (EPITHELIAL CELLS) - Place the bristle (nylon) head against the inside of the cheek. Pinch the cheek around the bristle head and firmly roll the swab around the pinched area. Pinch and swirl for about 15 seconds. Repeat this step with the remaining swab/s.

DRY THE SWABS - Allow the swabs to air dry for 3-5 minutes.

REINSERT DRY SWABS - After the swabs have dried, place them into the original packaging. Seal the packet closed with one of the barcode stickers.

Although buccal swab collection is a simple non-invasive collection procedure it is open to the likelihood of contamination if the procedure is not done correctly. Contamination from other sources is likely if the animal has been exposed to certain foods or bacterial contamination from any likely sources eg. tooth decay.

- Fill out all details on tamper proof bag
- Up to 6 separate samples can be placed into one bag. Swab packets may need to be folded over to ensure that they fit.
- Ensure you keep the receipt from the tamper proof bag for your records.



Protocols for collection are made available with all kits posted to clients and on the website. Key protocol requirements that are recommended to reduce possible sample contamination are:

- Try not to feed the animal for up to 20 minutes prior to collection.
- Samples can be collected from animals of any age. If collecting from suckling offspring, the offspring must be removed and isolated for at least 10 minutes.

Although human contamination does not affect the sample or the result, it is recommended to wear gloves when taking the sample. This is more necessary for veterinarians who are exposed to animals on a more frequent basis thus making cross contamination more of a possibility.

3.2 Semen samples can consist of semen straw or pellet. Used straws and/or bags that have stored semen and are also accepted. Semen can also be blotted directly onto blotting cards. Blotting cards are supplied free of charge upon request. Barcodes are allocated internally.

3.3 Blood samples should be collected in EDTA tubes (min 0.5ml) or EDTA blood can be spotted onto blotting cards. Blotting cards are supplied free of charge upon request. Barcodes are allocated internally.



The Process of DNA Collection Using Semen Samples (FAQs)

Overview

Unlike many other somatic cells, sperm DNA is very compact mainly due to replacement of histones (proteins) with protamines. Disulfide bonds formed within and between the protamines make the extraction of sperm DNA through standard techniques difficult when compared to other common material used for extraction e.g. Saliva or Blood. Furthermore, the spermatozoa themselves are protected by a membrane which is rich in disulphide bonds, making cell lysis very difficult.

Semen can be used to extract DNA and is a useful tool for assessing and identifying the semen – DNA profile (fingerprint) is unique and identifies that semen sample. Extracted DNA from the semen sample can also be used to run for hereditary diseases.

What do we need?

We can extract DNA from a semen straw or pellet. The straw or pellet can be shipped at room temperature – no liquid nitrogen is required. Let the sample thaw in the fridge and store at 4°C before shipping. Leaving the sample at room temperature for long periods can cause mould to appear on the sample. Place the specimen in a plastic zip lock bag – label the bag – and post as normal.



How much semen do you need?

Of course the more sample we have the more DNA we can extract. A full straw or breeding unit (pellet) gives the highest success rates and substantial quantity of DNA to allow you to screen for all diseases, traits and a DNA profile.

Any special conditions for shipping? What gives the best option of obtaining a result?

Option 1 (Preferred Option)

Place **the breeding unit (entire straw or pellet)** into a plastic zip lock bag (the smaller the better). An excellent way to ensure the straw is protected is to use the outside shaft of a ball point pen. Use the plastic top to seal one end. This shaft also helps protect the straw from any postal damage. Pellets are usually stored and shipped in an Eppendorf tube and this is sufficient for transporting. Or send 100ul (0.1ml) of semen in a microfuge tube, wrap or seal the lid with parafilm. This option has a 95% success rate.

Option 2 (second option)

Blot the semen straw or pipette the semen pellet directly onto a blotting card (filter paper). Blotting cards are available from Orivet free of charge and are preferred to normal filter paper. This tends to work well as “fresh is best” and therefore no issue of sample leaking or drying. This option has a 60-70% success rate.



Option 3 (least preferred option)

An **empty straw** can be used and in many cases is sufficient to get a DNA profile. To assist and ensure that the sample is successful simply place the empty straw in a small zip lock bag. Do not seal the ends with tape or other plugs. The straws are flushed out with sterile saline and this allows for any residual semen to be eluted into a tube.

One of the limitations of semen is the inability to store any sample surplus to the initial DNA testing requirements for future DNA testing of the animal. Since the amount of DNA extracted is limited we carry out DNA profiling (fingerprint) as a priority and will screen for diseases as a second option if there is sufficient DNA quantity available.

Please Note: that inhibitors in various extenders may impact on the ability to extract quality DNA. Since these vary from extender to extender we cannot provide 100% guarantee on obtaining quality DNA.

In some cases, only “partial profiles” can be obtained from a semen sample as this is sufficient for our laboratory to carry out screening and parentage confirmation. **A \$10 processing fee is included with any semen sample type submitted.**



Approved Collection Method

Sample submission checklist

- Ensure collection is done by a vet or Orivet Approved Collection Agent
- The microchip number is scanned + verified
- Pedigree cited (optional)
- The Approved Collection Method Form is completed + signed
- All details are provided on the tamper proof bag
- Tear the receipt off top of tamper proof bag for **your** records
- Client to order tests / activate their swabs themselves online
- Client to submit samples to Orivet*
(*unless vet or Collection Agent happy to send in to Orivet on client's behalf).





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