



DNA Bird Sexing

Technical White Paper

Determining a Bird's Sex

Knowing a bird's sex can be important from a number of health, breeding, commercial and ecological perspectives. For example, having an indication of the bird's sex can be important for veterinarian diagnosis of certain diseases, allow for more specialised sex-specific feeding plans to be used and to enable more reliable and successful commercial breeding and conservation activities.

Determination of sex in birds can often be difficult for many inexperienced bird owners and as such it often needs to be conducted by a local bird breeder or a veterinarian. Even then, determining bird's sex is not always straight forward or 100% accurate.

With certain species of birds, it is easy to determine the sex of individuals through sexual dimorphism. Sexually dimorphic species are characterised with distinct differences in appearance between male and female representatives; for example: colouration of the bird, overall body size, or specific structural features such as differences in beak sizes/shapes.



Above: Macaws are mostly monomorphic with little or no visual differences between the male (left) and female (right).



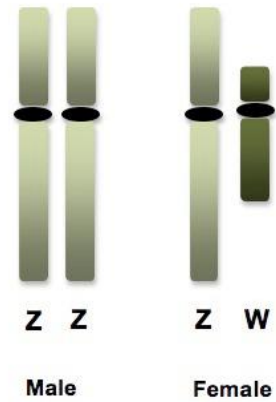
Above: The mallard duck (*Anas platyrhynchos*) is a dimorphic bird species, with distinct differences between the male (left) and female (right).

Nevertheless, dimorphic species (including species such as the pheasant and mallard duck) account for the minority of all bird species. Most birds are in fact monomorphic species, with no obvious visual differences in appearance between male and female birds, making it almost impossible to distinguish the sex based solely on their morphological traits. Furthermore, in certain dimorphic species (including parrots) mutations in beak/feather colouration and plumage patterns are no longer an accurate indicator of sexual dimorphism as a result of excessive in-breeding in the commercial bird trade. Finally, the chicks of dimorphic species can also be almost impossible to identify visually, for several weeks before they grow into their adult plumage. For example the sex of chicken chicks can only be visually determined 6-8 weeks after hatching.

Aside from visual determination of a bird's sex, other methods include behavioural observation, i.e. laying eggs, as only female individuals will do this. This method is very time consuming and often unreliable. Lastly a bird's sex can be investigated endoscopically. This expensive methodology requires bird to be put under anaesthetic to allow the veterinarian to insert the endoscope inside bird's abdominal air sacs, from which the entire abdominal cavity including reproductive organs can be observed. The veterinarian can then tell the sex of the bird by seeing either two internal testicles or one internal ovary. This method is highly invasive and can cause a large amount of distress to the bird.

DNA Based Bird Sexing

Bird sexing can now also be conducted using molecular methods by examining a bird's DNA to determine its sex. This method is very reliable and both time and cost effective. In birds, females have two different chromosomes (ZW), whereas males have both copies of the same chromosomes (ZZ). Using polymerase chain reaction (PCR) technique it is possible to investigate the differences between sex chromosomes and identify which chromosomes a bird has, therefore inferring the sex of a bird.



Collecting a Sample

Molecular bird sexing is minimally invasive and the least stressful method of sexing monomorphic or young birds. The DNA required for molecular sexing can be sourced from either freshly plucked feathers, blood or from an eggshell collected soon after hatching. Feathers are very easy to collect, and their collection will not cause any harm to the bird, however, multiple feathers will be required to obtain enough DNA for analysis. Blood can be obtained from clipping the nail of the bird slightly higher than usual during regular nail grooming. Blood collection is more invasive, but it is a better source of DNA. Lastly, eggshell collected straight after hatching is a great source of DNA material and its collection is non-invasive and non-stressful to the bird.





New Bird Sexing Service

At SureScreen Scientifics, we now offer a bird sexing service. By running a very detailed optimisation of the method in our lab we have narrowed down criteria for feather collection in order to maximise the DNA yield. During collection, feathers need to be freshly plucked in order to provide us with enough DNA material for analysis. The DNA is only present in the very tip of the feather quill, therefore plucking the feathers is the only way to guarantee a reliable source of DNA. Moulted feathers cannot be used as they will not have enough DNA for a reliable sex determination to be conducted. Feathers need to be no more than 5 days old when received in our lab.

Using feathers' quills the DNA is extracted and processed using the PCR method to identify and amplify a region of DNA within the sex chromosomes. Using a process known as gel electrophoresis we can then visualise this DNA, which will present either 2 bands of DNA (indicating two different chromosomes: ZW) for females or 1 band of DNA (indicating two copies of the same sex chromosome: ZZ) for males. Using these definitive results, we can accurately determine the sex of the bird species.

This approach is a widely universal method for bird sexing and has been tested on over 80 species of birds including species of parrots, owls, pigeons, chickens, storks, woodpeckers and many more.



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