

genetic testing  
**CAREL TESELING**



## NEW TEST IDENTIFIES BLACK WAGYU CONTENT

The Australian Wagyu Association (AWA) has developed a genomic test to determine the level of Japanese Black (Black Wagyu) in non-pedigreed crossbred animals. This ground-breaking development, known as the Crossbred Wagyu Test (CWT) will help the supply chain determine feeding regimes and the propensity of cattle to produce a genuine Wagyu eating experience.

AWA's Carel Teseling explains the purposes of the test, how it works and delves into the prefectural nuances of the breed in Japan.

### PURPOSE

- » To assist the beef industry in identifying the approximate level of Black Wagyu content from a non-pedigree crossbred animal's DNA sample.
- » Assist in selecting a new class of breeding animals to purchase or to register with the AWA. Animals with tested Black Wagyu content can be registered into the Wagyu Content Register and used for breeding purposes.
- » The AWA board is reviewing the Bylaws to determine the appropriate level at which animals with tested Black Wagyu content can enter the Herdbook as a Purebred animal.
- » Supply chain verification of non-pedigree, crossbred animals. Testing may be particularly valuable before feedlot entry, offering a new cost-effective means for producers, cattle buyers, and processors to determine the Black Wagyu content in commercial crossbred cattle mobs. The results could, for instance, facilitate categorising crossbreds into the progeny of the Black Wagyu bull or a mickey bull. This knowledge would significantly influence the choice of optimum feeding regimes, processing, and marketing.

### INTRODUCTION

The Australian Wagyu Association has developed a genomic test to determine the level of Japanese Black (Black Wagyu) content in a DNA sample from a crossbred animal. The test measures Black Wagyu content in terms of the animal's relationship to the mainstream Black Wagyu sub-population available in Australia. While some Australian fullblood animals will return lower CWT values, this needs to be recognised as an indication of their importance as "outliers" within the local gene pool, and the valuable contribution they can make to maintaining genetic diversity and providing future selection flexibility. Further development work will endeavour to measure better and define these outlier populations, which have already been identified as descendants of specific, original Japanese Black prefectural herds.

### HOW THE TEST WORKS

Essentially the CWT measures the genomic "distance" between the genotype of a particular animal being tested and the "clouds" or reference sets of genotypes available for other breeds and delivers the estimated breed content level of each breed within that animal.

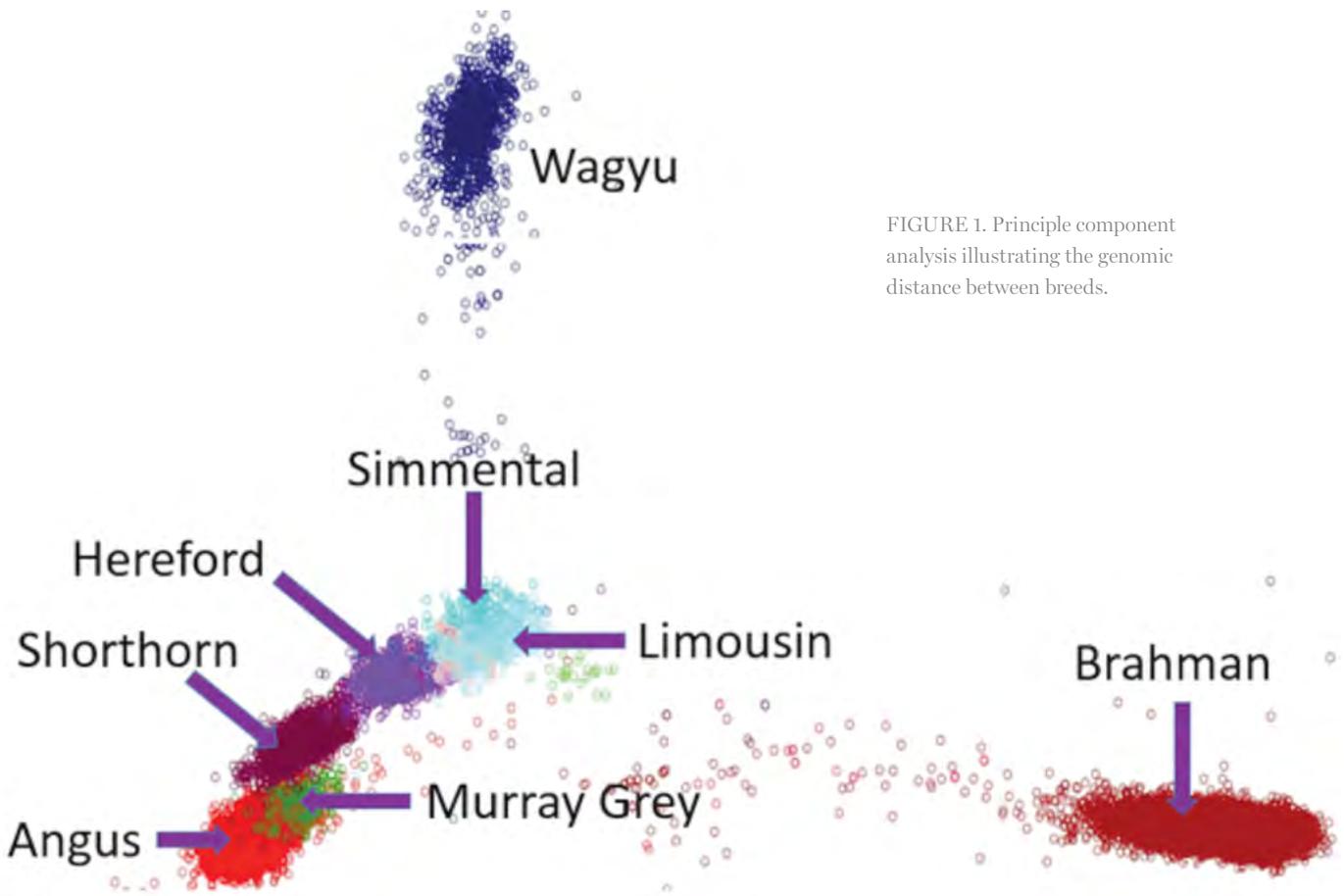


FIGURE 1. Principle component analysis illustrating the genomic distance between breeds.

The CWT software has been developed by the Animal Genetics & Breeding Unit (AGBU) at the University of New England. It uses a reference dataset for each breed type whose genotypes are currently available. At least 100 individual animal genotypes, representing the genetic variation in the breed, are required to form an accurate representation of a particular breed, so only the major breeds have sufficient genotypes to enable them to be included at this stage. Each animal in the reference set requires a genotype of sufficient SNP (Single Nucleotide Polymorphism) density to enable accurate breed determination, with the Low-Density GeneSeek Genomic Profiler (GGPLD) of some 34,000 SNPs currently the minimum (previous GGPLD SNP levels were 10K, then 20K and these have also been used). The breed genotypes have been provided by a range of industry contributors including AWA through its Wagyu Collaborative Genetics Research Project, Meat & Livestock Australia through the various Beef Cooperative Research Centre projects and other breed associations.

The CWT uses a two-step process to calculate the breed content:

- » Firstly, the software calculates the unique allele frequencies for each breed in the reference dataset. An allele is one of a pair of genes that appear at a particular location on a particular chromosome and control the same characteristic, such as blood type or coat colour.
- » Secondly, it calculates the correlations between the allele frequencies of the test animal's genotype and those of the other breeds, and this represents the breed percentages.

### BREEDS IN THE REFERENCE DATASET

The reference dataset currently has more than 10,000 genotypes representing 11 of the most prominent Australian beef breeds and consists of:

- » Approximately 1,600 Wagyu and Crossbred Wagyu genotypes ranging from Fullblood Wagyu 100% (almost exclusively Japanese Black) to Crossbred Wagyu F1 50% provided by AWA derived from its Wagyu Collaborative Genetics Research Project co-funded through the MLA Donor Company.
- » Approximately 7,000 genotypes from 10 other beef breeds largely provided from the Beef Co-operative Research Centres (Beef CRC's) I, II and III co-funded by the beef industry and the MLA Donor Company.
- » The 11 breeds currently having genotypes in the reference dataset are Angus, Brahman, Charolais, Droughtmaster, Hereford, Limousin, Murray Grey, Santa Gertrudis, Simmental, Shorthorn and Wagyu.

This reference dataset of the various breeds in their genomic "clouds" is graphically represented in Figure 1 (above), showing the distance between each breed "cloud".

### THE WAGYU REFERENCE DATASET

The term Wagyu is a general term for about five breeds of cattle bred in Japan. The Japanese word is 'our' (wa) and 'cattle' (gyu). The CWT measures only content from the dominant >>>

<<<

Japanese Black breed. There are three major Japanese Black bloodlines well represented in the Australian sub-population:

- » Kumanami and Tajima from the Hyogo Prefecture
- » Itozakura & Fujiyoshi from the Shimane Prefecture
- » Kedaka and Eikou from the Tottori Prefecture

Two major red (Japanese Brown) sub-populations (Akaushi) are also represented in Australia, but are not covered by the CWT. These are:

- » Kochi; and
- » Kumamoto

Modern Wagyu cattle breeds are the result of crossing of the native cattle in Japan with imported breeds. Systematic crossing began in 1868 during the Meiji restoration. Brown Swiss, Devon, Shorthorn, Simmental, Angus, Ayrshire and Korean cattle were imported during this period. The infusions of these British, European and Asian breeds ceased about 1910. However, it can be expected that some evidence of these breeds still exists in some prefectural Fullblood Wagyu cattle herds in Japan. The red population was strongly influenced by Korean and European breeds, particularly Simmental.

The Australian herd is descended from a foundation Black Wagyu population exported to the USA and Australia from Japan between 1976 and 2001. This comprised some 300 animals.

### THE CROSSBRED WAGYU TEST RESULTS FULLBLOOD RESULTS

An animal is regarded as a Fullblood Wagyu whose forebears originate from Japan. Registered Fullbloods require DNA parent verification to both the sire and dam to ensure complete pedigree accuracy. Over 300 registered Black Fullblood Wagyu were represented in the reference dataset with their test results shown in figure 2 (right).

The results highlight the genetic diversity available in the registered black Australian Fullblood population. The Fullblood animals with lower CWT results are highly valuable to the future of the Black Wagyu

### NUMBER OF ANIMALS

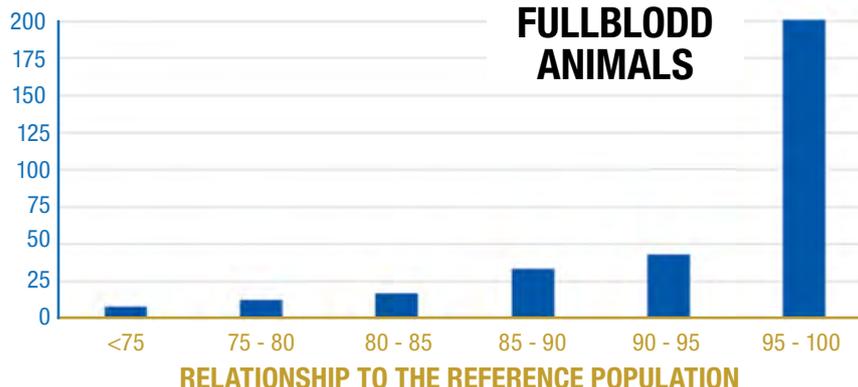


FIGURE 2. Frequency distribution of registered black Fullblood animals.

### NUMBER OF ANIMALS

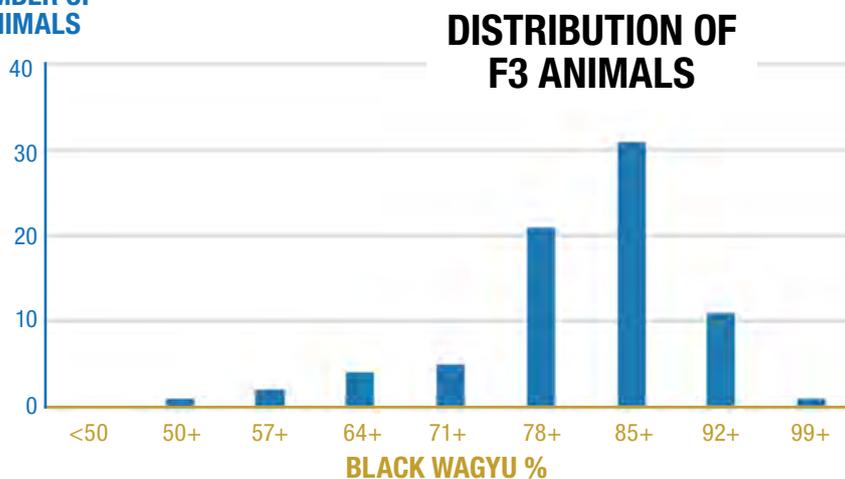


FIGURE 3. Frequency distribution of F3 Wagyu cross animals.

population in Australia as they provide the only current opportunities available to retain genetic diversity and deliver enhanced selection options for traits such as superior conformation, growth and maternal capability, with retained marbling.

### CROSSBRED RESULTS

Initially, it was expected that the Black Wagyu content in crossbred Wagyu animals would follow the traditional genetic norm of:

- » F1 or first Wagyu cross delivering 50% Wagyu
- » F2 or second cross Wagyu delivering 75% Wagyu
- » F3 or third cross Wagyu delivering 87% Wagyu
- » F4 or Purebred Wagyu delivering 93% Wagyu

However, the genomic variation identified in the genotyped registered Black Fullblood animals was reflected in their crossbred progeny resulting in a range for any given crossbred grade as shown in figure 3 (above).

**NOTE:** It is expected that F3 animals with higher than 92% were out of cows that already had some Black Wagyu influence.

Therefore, crossbred progeny of a high scoring Black Fullblood sire can be expected to have content close to the expected genetic norm, while progeny from sires with other prefectural genomic content may show less than the expected genetic norm. Providing the sire is registered with known performance through its Estimated Breeding Values (EBVs), this will not pose a problem and indeed may be a distinct advantage if the sire has higher growth and other attributes.

## HOW THE CWT MAY BE USED WAGYU CONTENT REGISTER

There are many animals with Wagyu content in member herds where pedigree cannot be proven through DNA parent verification and so cannot be registered as Fullbloods or Purebreds. However, they are of considerable value to the Wagyu gene pool. These animals can now be registered in the Wagyu Content Register and their performance data input for analysis through Wagyu BREEDPLAN. The Crossbred Wagyu Test can provide the Black Wagyu breed percentage required by the Content Register Bylaws:

### CONTENT REGISTER

1. The Content Register makes provision for the registration of Wagyu influenced animals not eligible for registration in the fullblood or purebred registers.
2. It shall be a requirement for the registration of a Content Register calf that its sire has a DNA profile recorded with AWA at a minimum level to be specified by AWA.
3. Prior to registration a hair sample of the animal will be provided to AWA.
4. Parent verification of calves will not be required.
5. Random parent verification of calves will be conducted at a frequency determined by the board at no cost to the animal owners. Animals found to have an incorrect sire will be deregistered if the correct sire is not identified by the owner at the owner's cost.
6. If no Wagyu content is claimed for a base animal, the breed (or breed combination) of the base animal shall be recorded.
7. **If Wagyu content is claimed for a base animal, a breed content test will be required for the animal. Animals registered in the content register shall be assigned a percentage breed content calculated from the percentage Wagyu of the sire and dam or results from the Crossbred Wagyu Test.**

Registration of animals with Wagyu content is expected to significantly increase

the animal's sale and therefore capital value as buyers can have greater confidence in the claimed Wagyu content.

### TESTING ANIMALS WITH THE CWT

An animal can be tested to determine its Black Wagyu content using the Crossbred Wagyu Test. This may be of value in:

- » Assisting in selecting breeding animals to purchase or to register with the AWA. Animals with tested Black Wagyu content can be registered in the Wagyu Content Register and used for breeding purposes.
- » Upgrading an animal into the Herdbook. The AWA board is reviewing the Bylaws to determine the appropriate level at which animals with tested Wagyu content can enter the Herdbook as a Purebred animal.
- » Supply chain verification of animals. Testing may be particularly valuable before feedlot entry, offering a new cost-effective means for producers, cattle buyers, and processors to determine the Black Wagyu content in commercial crossbred cattle mobs. The results could, for instance, facilitate categorising crossbreds into the progeny of the Black Wagyu bull or a mickey bull. This knowledge would significantly influence the choice of optimum feeding regimes, processing, and marketing.

### FURTHER INFORMATION

For further information see the Frequently Asked Questions or contact

Carel Teseling  
AWA Technical Services Manager

☎ 02 8880 7703  
0439 368 283

✉ [carel@wagyu.org.au](mailto:carel@wagyu.org.au)

