

Inheritance of different coat colours in Newfoundland dogs in Poland

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The aim of the study was to present the manner in which coat colour genes are inherited in the Newfoundland dog breed and to estimate the number of dogs with various coat colours in the Polish Newfoundland dog population in 2017. This population numbered 656 dogs, including 248 males and 408 females. The estimated number of dogs of this breed also included all registered puppies, broken down by gender and coat colour. The genes determining coat colour are described, including more precisely the genes responsible for the coat colour of the Newfoundland breed. According to FCI regulations, the coat colours for Newfoundland dogs are black, brown and black-and-white. Other colours, such as brown-and-white or grey, are not recognized for breeding purposes in Europe. The study found that the dominant black coat was predominant in the Polish Newfoundland dog population in 2017. These dogs could be heterozygous at some other loci and have undesirable alleles. The second most common coat colour was brown, while the fewest dogs had spotted coats. The group with spotted coats contained more males than females, in contrast to the other two colour variants. There were also individuals with the grey coat colour, which is not accepted for breeding, as the result of mating of parents with proper coat colours. An understanding of how dog coat colours are inherited and the need for tests to determine coat colour genotypes would make it possible to foresee the occurrence of incorrect colours in subsequent generations, which is crucial for Newfoundland dog breeders, whose goal is to obtain dogs whose coat colour is in line with the FCI standard.

KEY WORDS: dog / inheritance / coat colour / Newfoundland dog

The Newfoundland dog breed arose in North America. There are two theories regarding its origin. According to the first, large, black bear dogs were brought by Vikings to the area that is now Newfoundland. These dogs crossed with native breeds, but the colour black was clearly dominant in the offspring. According to the second theory, Newfoundland dogs originally came from Asia through Alaska and were of the Tibetan mastiff type. The

changes in the type and its establishment continued over a period of about five centuries. The spotted coat in this breed was introduced at the beginning of the 18th century, with the arrival of English settlers on the American continent, who brought with them large black-and-white and brow-and-white dogs. They crossbred with the black, massive native dogs, giving rise to today's spotted coat in this breed [5, 11].

According to the FCI standard, only black, brown and black-and-white dogs may be bred in Europe, and thus in Poland, whereas in the United States, grey individuals are accepted as well. Males and females accepted for breeding must meet the requirements of the breed standard, one of the most important features of which is coat colour. Any colour other than those mentioned above disqualifies the animal for breeding. This applies to imported animals as well. However, breeders import dogs or their semen not only from European countries, but also from the United States. Recently, puppies with incorrect colour born of correctly coloured parents have begun to appear in Polish kennels. Therefore the aim of the study was to describe the inheritance of coat colour in Newfoundland dogs and to estimate the number of dogs of individual colour variants in this breed in Poland, including variants not recognized for breeding.

Material and methods

To estimate the number of Newfoundland dogs in the Polish population, which was considered to be representative of kennel clubs in the FCI, we collected data from the websites of licensed kennels of this breed that possess and show dogs bred in Poland and dogs imported but used and kept in Poland, as well as puppies born in Poland in 2017.

The data include the sex and coat colour of the parents and additionally the number of puppies born in 2017, along with their colours. The colour of the individuals was divided into categories:

- black (a white patch on the breast did not exclude the dog from this category)
- brown
- spotted
- grey

The study took into account the colour grey, which is not recognized by the FCI but has appeared in many Newfoundland kennels in Poland. Individuals that were mainly black, but had white spots not only on the breast, were included in a separate colour category as 'spotted'.

Additionally, to supplement the data on the number of dogs of each colour variant (in case the data on the websites were incomplete), a questionnaire with questions regarding the coat colours of animals at kennels was prepared and posted on the website of the popular Newfoundland dog breeders forum NOWOFUNDLAND.PL.

In total, the coat colour of 656 dogs was analysed. In the case of 623 individuals, information on colour and gender were obtained from kennel websites, and in 33 cases from the questionnaires. The collected data were presented numerically and as percentages, broken down by sex, age and colour.

Results and discussion

Data obtained from breeding websites, additionally supplemented with survey results, were used to summarize the numbers of Newfoundland dogs, broken down by sex and coat colour. The results are shown in Table 1.

Table 1

Estimated number of Newfoundland dogs in Poland in 2017 year by sex and coat colour

Coat colour	Males	Females	Total
Black	167	318	485
Brown	45	61	106
Spotted	32	27	59
Grey	4	2	6

In 2017, there were 656 Newfoundland dogs in Poland, including 248 males, which was 38% of the studied population, and 408 females, constituting 62% of the population. The estimated population of Newfoundland dogs given in Table 1 also includes all puppies born and registered in 2017.

The predominant colour was black, which was noted in 74% of the population. Black males accounted for 25.5% of the Newfoundland population, and black females for nearly half (48.5%) of all registered individuals. Brown dogs comprised 16.2% of the total population, of which males accounted for 6.9% and females 9.3%. Spotted individuals made up 9% of the population – males 4.9% and females 4.1%. Additionally, four males (0.6%) and two females (0.3%) had a grey coat (unrecognized colour), for a total of six individuals (Table 1), which was less than 1% of the total population (0.9%).

According to the Main Breeding Committee of the General Board of the Polish Kennel Club, which keeps breed statistics published in the quarterly *Pies*, the population of the Newfoundland breed in 2016 was 606 individuals, including 225 puppies [20], and thus in 2017 the number had increased by 50. In 2017, there were 128 adult Newfoundland males, and 120 male puppies were born. In the same year, there were more than twice as

many adult females as males, i.e. 284, while only half as many females were born that year – 141. In percentages, adult females constituted 43.3% of the Newfoundland population, female puppies 21.6%, adult males 20%, and male puppies 18.3%. Table 2 presents the research material broken down by sex, age and colour.

In 2017, among black Newfoundland dogs, adult males accounted for 13.6% of the total Newfoundland population and male puppies for 12%. Black adult females comprised 34.5% of the total population, while black female puppies accounted for only 14% (Table 2).

Among brown-coated individuals, there were almost the same number of adults and puppies in the case of both sexes. Brown adult males comprised 3.5% of the population, and brown male puppies 3.4%. In the case of females, brown adult females accounted for 4.6% of the population, and brown puppies for 4.7% (Table 2).

Spotted adult males constituted 2.1% of the research material and spotted male puppies 2.7%. Spotted adult females comprised 5.9%, while female puppies of this colour accounted for only 2.7% (Table 2).

Adults were predominant among grey animals. Two adult males (0.3%) and the same number of male puppies (0.3%) had this coat colour. There were also two (0.3%) adult grey females (Table 2).

The results presented above indicate that black was the most common colour in the Newfoundland breed (Table 1 and 2). This is the colour that originally appeared in this breed. Brown Newfoundland individuals are less common than black ones (Table 1 and 2), but this colour is also recognized, and buyers readily purchase dogs with this coat colour. In black and brown dogs, a small white spot is allowed on chest and the tips of the paws. Pigment cells, i.e. melanocytes, are believed to reach these sites on the dog's body last during ontogenesis, and sometimes they do not reach them at all [5]. For this reason, white markings are not considered a coat defect or as spotting in the Newfoundland breed.

Table 2

Estimated number of Newfoundland dogs in Poland in 2017 year by age, sex and coat colour

Coat colour	Sex and age of animal			
	males		females	
	adult	pups	adult	pups
Black	89	78	226	92
Brown	23	22	30	31
Spotted	14	18	26	18
Grey	2	2	2	0

In analysing the genotype that determines the dog's coat colour, the genes that make up the coat genotype can be divided into two groups. The first are genes indirectly involved in producing a given colour, while the other comprises genes directly responsible for the visible colour. In the Newfoundland breed, genes in the first group are from the **C**, **A**, **E**, **G**, **H**, **I** and **M** loci, while genes from the **K**, **B**, **D** and **S** loci directly influence the coat colour. The influence of genes from the **T** and **R** loci on the coat of Newfoundland dogs remains unknown.

Table 3 lists the possible genotypes and phenotypes of Newfoundland dogs of different colours, including those that are not recognized by the FCI. However, the set of alleles in the **S** locus resulting in the **s^Ps^P** genotype was not included, because there are no individuals whose spotting is so extensive that they are nearly white. Gene arrangements at the **B** and **D** loci in which animals are carriers of recessive genes are underlined.

The **TYR** gene at the **C** locus is responsible for producing a melanin precursor. Most dogs are dominant homozygotes at this locus and mutations have been found in only a few breeds. There is also a recessive albinism gene at this locus in many animal species, but thus far it has not been detected in dogs [16]. As there are no extremely light (albino) individuals in the breed, it should be assumed that at the **C** locus all individuals will be dominant **CC** homozygotes.

Table 3

Possible genotypes and phenotypes and observed coat colours in Newfoundland dogs

Genotype	Phenotype	Remarks
K^BK^B BB DD SS K^BK^B BB <u>Dd</u> SS K^BK^B <u>Bb</u> DD SS K^BK^B <u>Bb</u> <u>Dd</u> SS	black	coat colour accepted for breeding, could be mated with black and black spotted animals
K^BK^B bb DD SS K^BK^B bb <u>Dd</u> SS	brown	coat colour accepted for breeding, could be mated with black animals
K^BK^B BB dd SS K^BK^B <u>Bb</u> dd SS	grey	coat colour not accepted for breeding
K^BK^B BB DD Ss^P K^BK^B BB <u>Dd</u> Ss^P K^BK^B <u>Bb</u> DD Ss^P K^BK^B <u>Bb</u> <u>Dd</u> Ss^P	black spotted	coat colour accepted for breeding, could be mated with black animals
K^BK^B bb dd SS K^BK^B bb dd Ss^P K^BK^B bb <u>Dd</u> Ss^P	cream cream spotted brown spotted	
K^BK^B bb DD Ss^P	brown spotted	coat colours not accepted for breeding
K^BK^B BB dd Ss^P K^BK^B <u>Bb</u> dd Ss^P	grey spotted grey spotted	

The *ASIP* gene located at the *A* locus is responsible for the synthesis and distribution of two pigments, black eumelanin and yellow/red pheomelanin. The zonal distribution of the two pigments in both the hair and the skin is determined by a series of multiple alleles $A^y > a^w > a^t > a$ [10, 18]. Without performing a genetic test, it is not possible to conclusively state which alleles a Newfoundland individual will have at the *A* locus. Their potential effect is associated with the *K* locus, in which Newfoundland dogs are dominant $K^B K^B$ homozygotes, and thus an epistatic effect of this gene arrangement in relation to the *A* locus is manifested, preventing them from exerting their effect.

The gene responsible for the base coat colour in the Newfoundland breed is *CBD103* at locus *K*, which is the site of the multiple allelic series $K^B > k^{br} > k^y$ [9]. The K^B gene determines the appearance of the colour black dominating in an interaction with the genes at locus *E*, and is epistatic to the genes from locus *A*. This locus is the site of gene k^{br} , which determines stripes in dogs and is epistatic to the A^y gene [15, 18]. Observations of the appearance of dogs indicate that black Newfoundland dogs will be, as mentioned above, dominant $K^B K^B$ homozygotes. Moreover, this breed has no dogs with a striped coat (effect of the k^{br} gene) or any other coat determined by genes at the *A* locus, which would be manifested in the absence of the K^B gene (our own observations; conversation with breeders). If individuals with a yellow coat were to appear in this breed, it might indicate the presence of the k^y allele in the breed. For example, striped or yellow puppies could be born of two black $K^B k^{br}$ or $K^B k^y$ heterozygous individuals. It should also be taken into account that the *K* locus is epistatic to the *A* locus, so in a potential homozygous $k^y k^y$ arrangement, puppies could also have other coat colours determined by *A* locus genes, namely the wild-type (wolf pattern), black-and-tan, or recessive black coat. These colours, however, do not appear in Newfoundland dogs, which confirms the assumption that they are dominant $K^B K^B$ homozygotes.

The next gene responsible for the coat colour of Newfoundland dogs is *MC1R* at the *E* locus, in which there is also a multiple allelic series determining the production of black (eumelanin) and yellow/red (pheomelanin) pigment. The interaction of the genes from the *A* and *B* loci leads to the appearance of a melanistic mask and other types of masks in various dog breeds. In the recessive *ee* homozygote, the yellow/red pigment (pheomelanin) is found in the hair, while black eumelanin is produced in the skin [15, 18]. At the *E* locus, Newfoundland dogs seem to be dominant *EE* homozygotes (there have been no genetic tests). They do not possess E^M alleles responsible for a melanistic mask. Their coat colour is also not lightened by an *ee* allelic arrangement, with a black nose and mucous membranes.

Another gene influencing the coat colour of Newfoundland dogs is a gene at the *B* locus. Here, Newfoundland dogs with a black coat can be dominant *BB* homozygotes or *Bb* heterozygotes. They can also be recessive *bb* homozygotes and will be phenotypically brown, including a brown nose and mucous membranes. The occurrence of four alleles has been described at this locus: *B*, *b^s*, *b^c* and *b^d*. Alleles *b^s*, *b^d* and *b^c* in a homozygous arrangement are known to determine the same phenotypically indistinguishable brown coat colour. They are found in different combinations in different breeds [13, 15, 18]. However, it is unknown which of them are found in Newfoundland dogs.

The next group comprises genes responsible for lightening of the base coat colour. The first of these is *MLPH* from the **D** locus, which in the recessive homozygote lightens eumelanin and pheomelanin in the hair and skin, changing black to grey and brown to cream-coloured [1, 4, 14, 19]. At the **D** locus, as in the case of the **B** locus, dogs of this breed can be dominant **DD** homozygotes or **Dd** heterozygotes. Their phenotype will then be determined by alleles from the **K** and **B** loci, i.e. they will be black or brown. If at the **D** locus the Newfoundland dog is a recessive **dd** homozygote, its coat will be lightened to grey. The eyes, nose and mucous membranes will also be lightened to grey. This coat colour, although not recognized for breeding, may appear following mating of two black individuals that are **Dd** heterozygotes. In this case puppies with grey, defective coats can be born of two properly coloured parents (Table 3).

The group of genes determining a spotted coat includes *SILV* at the **M** locus, which causes the merle coat and only acts at the site where dark eumelanin occurs. The merle gene is lethal in a homozygous arrangement (**MM**), while heterozygous individuals (**Mm**) survive, but suffer from eye disorders caused by this gene. The merle coat is characteristic only of a few sheepdog breeds and is very rare in other breeds [3, 6]. Another gene, *PSMB7* at locus **H**, which as a dominant gene occurs only in Harlequin Great Danes, interacts with the **M** locus, lightening the grey merle background to white [2]. At the **H** locus, Newfoundland dogs are also recessive **hh** homozygotes, as the dominant **H** allele occurs only in Great Danes. There is also no merle coat observed in this breed, so at the **M** locus Newfoundland dogs will be recessive **mm** homozygotes.

Incomplete dominance occurs at the **G** locus, and heterozygous individuals turn grey later than dominant homozygotes, while recessive individuals retain their intense colour [15]. At this locus Newfoundland dogs are recessive **gg** homozygotes, as progressive greying is not observed in this breed. In the next, hypothetical locus **I** they can be recessive **ii** homozygotes, while the cream coat is probably caused by a gene from the **D** locus.

That final gene influencing the coat colour of Newfoundland dogs is *MITF1* from the **S** (spotting) locus. The multiple allelic series **S**, **sⁱ**, **s^p**, **s^w** is believed to occur at this locus [12, 15, 17]. Research is continually being conducted on this locus, but only two alleles from the series, **S** and **s^p**, have been confirmed [7, 8]. There is incomplete dominance at this locus, which means that dominant **SS** homozygotes will not have spots, while **Ss^p** heterozygotes will manifest an intermediate form of spotting. The genes responsible for the distribution of spots are unknown. Recessive homozygous **s^ps^p** individuals will be very extensively spotted (nearly white). This coat colour is not found among Newfoundland dogs in Poland and was not taken into account in the present study. It is not known which genes determine the distribution of white spots, but they may be modifier genes [5]. Newfoundland dogs have white spots on the muzzle, a white blaze on the head, a white chest, white feet, and a white tail tip. This is known as Irish spotting. There can be more white spots, but there must be a black saddle on the back and a black head.

There are two more hypothetical genes at the **T** and **R** loci that can slightly modify the black-and-white coat. The **TT** allele arrangement causes the appearance of flecks on the background of the white areas, so phenotypically it will only be manifested in black-and-white individuals. There is incomplete dominance at this locus and Newfoundland dogs are probably **Tt** heterozygotes here, because the flecks, also known as ticking, are present in small numbers, usually on the muzzle and forelegs. Spotted dogs with pure white spots may be recessive **tt** homozygotes. The final hypothetical gene is located at the **R** locus, and in the heterozygous or dominant homozygous arrangement it is responsible for roan colour. Here there is incomplete allelic dominance, as various degrees of this trait are observed. Roan colour is manifested as a mixture of the base coat hairs with white ones, and is only found at the site of white spots. Therefore we can only speak of roan colour or phenotypically visible expression of the roan gene in the case of black-and-white individuals. In the Newfoundland breed there are also individuals born with other, unrecognized coat colours. These are presented in Table 4, but they will not be discussed further because they are not currently found in the Polish Newfoundland population.

Table 4 presents possible mating variants in Newfoundland dogs and their effects in the form of puppies with various coat colours recognized for breeding.

Table 4 describes the case of mating of two spotted Newfoundland dogs, which can produce puppies with extensive white spotting (mating variant no. 6). There were not many dogs and puppies of this type, possibly because breeders rarely mate two spotted individuals, believing them to be anatomically weaker and to have a lighter skeleton and lower quality hair (our own observations; conversation with breeders). What is more, brown puppies can be born to two black individuals, when both parents are **Bb** heterozygotes at the **B** locus (mating variant no. 3). If one of the parents additionally carries the spotting gene, a spotted brown puppy may be born, which will also not be accepted for breeding due to its coat colour. The same situation can occur when grey puppies are born to two black parents that are both heterozygous at the **D** locus. This case was not included in Table 4 because the grey coat colour is not accepted for breeding. However, it was taken into account in the estimation of numbers, because such puppies are now born in Poland (our own and survey information). It can be surmised that their numbers will increase, because this coat colour is approved for breeding in the United States, so recessive **d** alleles will be present in the Newfoundland dog population. Dogs of this breed are imported to Poland, including from the United States, and increasingly semen for insemination is imported as well (also from the US), which may be the reason for the growing frequency of the grey coat colour in Poland.

The roan coat (**R** allele) and ticking (**T** allele) are also not entirely desirable. Black ticking on a white background appears most often on the white spot on the muzzle and on the feet. Roan colouring, on the other hand, occurs most often in at the edges of spots, resulting in a kind of unclean combination of the base colour with the white spot. Roan colour and ticking may also appear on other areas of the body, but it will not appear on the black hair. Breeders try to eliminate the dominant form of these alleles.

Table 4

Results of mating of parents with different coat colours accepted for breeding in Newfoundland dogs

No	Genotype of sire	Genotype of dam	Possible genotypes and phenotypes of progeny
1.	K^BK^BBBSS (black)	K^BK^BBBSS (black)	K^BK^BBBSS (black)
2.	K^BK^BBBSS (black)	K^BK^BBbSS (black)	K^BK^BBBSS (black) K^BK^BBbSS (black)
3.	K^BK^BBbSS (black)	K^BK^BBbSS (black)	K^BK^BBBSS (black) K^BK^BBbSS (black) K^BK^BBbSS (black) K^BK^BbbSS (brown)
4.	K^BK^BBBSS (black)	K^BK^BBBSS^p (black spotted)	K^BK^BBBSS (black) K^BK^BBBSS^p (black spotted)
5.	K^BK^BBBSS (black)	K^BK^BBbSS^p (black spotted)	K^BK^BBBSS (black) K^BK^BBbSS (black) K^BK^BBbSS^p (black spotted) K^BK^BBBSS^p (black spotted)
6.	K^BK^BBBSS^p (black spotted)	K^BK^BBBSS^p (black spotted)	K^BK^BBBSS (black) K^BK^BBBSS^p (black spotted) K^BK^BBBSS^p (black spotted) K^BK^BBBSS^p (black extensively spotted)
7.	K^BK^BBBSS (black)	K^BK^BbbSS (brown)	K^BK^BBbSS (black)
8.	K^BK^BbbSS (brown)	K^BK^BbbSS (brown)	K^BK^BbbSS (brown)

Other unrecognized colours may also occur in the breed. They appear after mating of dogs with a recognized coat colour, but which are heterozygous for certain genes. Table 5 gives examples of mating variants that could result in unrecognized coat colouring in Newfoundland dogs.

Summing up, in 2017 there were an estimated 656 Newfoundland dogs in Poland, including 261 puppies, which is an increase of 50 individuals compared to the previous year. Black dogs were the most numerous in this group (485 individuals), because this colour is the most recognizable for the breed, very popular, and good in terms of conformation. As many as 66% of dogs with this colour variant in Poland were female. The second most common variant was the brown coat (106 individuals). There is an upward trend in this colour in Newfoundland dogs, because breeders and potential buyers consider these dogs to have an impressive appearance. The spotted coat was the least common of the colours accepted for breeding in this breed (59 animals). As mentioned above, dogs with this colouring have a lighter skeleton (our own observation) and are the least readily chosen for

Table 5

Incorrectly coloured offspring from mating of two dogs acceptable for breeding (only genotypes of incorrectly coloured puppies are shown here)

No	Genotype of sire	Genotype of dam	Possible genotypes and phenotypes of progeny
1.	K^BK^BBbDdSS (black)	K^BK^BBbDdSS (black)	K^BK^BbbddSS (cream) K^BK^BBBddSS (grey)
2.	K^BK^BBbDdSS (black)	K^BK^BBbDdSs^p (black spotted)	K^BK^BbbddSs^p (cream spotted) K^BK^BBBddSs^p (grey spotted)
3.	K^BK^BBbDDSS (black)	K^BK^BBbDDs^p (black spotted)	K^BK^BbbDDs^p (brown spotted)
4.	K^BK^BBBdDSS (black)	K^BK^BBBdDs^p (black spotted)	K^BK^BBBddSS (grey) K^BK^BBBddSs^p (grey spotted)
5.	K^BK^BBBdDSS (black)	K^BK^BBBDDSS (black)	K^BK^BBBddSS (grey)
6.	K^BK^BbbDdSS (brown)	K^BK^BbbSS (brown)	K^BK^BbbddSS (cream)

further breeding. In order to increase diversity within the Newfoundland breed, dogs are imported from abroad (including the United States), which results in the appearance in Poland of an increasing number of individuals with a grey coat colour, which is not accepted in Europe but is recognized in the USA.

It seems interesting and important to monitor future changes in the numbers of Newfoundland dogs in Poland, especially the frequency of individual colour variants recognized in breeding, as well as the appearance of undesirable coat colours in this breed. It is also important to educate breeders of Newfoundland dogs regarding the inheritance of their dogs' coat colour and the need to perform colour tests on individuals they plan to purchase or whose semen they plan to import, especially in the case of American dogs.

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