Reds, Creams and Inbetweens

Dr Kerry Fowler MSc PhD Grad Dip Ed

Published in NSW CFA Catching Up 5(3/4):5-6(2003)

Background

Current registration systems in most Australian cat councils work on the concept that two solid cats cannot produce a tabby kitten. In the main this policy is absolutely correct however in the case of the orange or 'O' gene cats ie creams, reds and the less vibrant reds known as 'gingers' and 'marmalades', sticking to this hard and fast rule often results in no end of controversy and confusion. Breeders are obliged to register red or cream tabby marked kittens from solid parents as red or cream selfs and in turn judges are required to assess them as such despite some of these red or cream kittens and cats having an abundance of clear tabby markings. Being a sex-linked gene, the 'O' gene coat colour issues extend to tortoiseshells, blue-creams and tricolours. This article identifies some of the misunderstanding surrounding breeding and judging 'O' gene cats including cameos and apricots, and draws on current genetic knowledge, registration policies and show standards from other councils to help resolve some of the re-occurring questions.

Genetic principles

In order to help put some of the debate and perplexity aside it is worth keeping in mind that there are four basic genetic rules that help us to understand the appearance of red or cream tabbies and solids. First, the tabby pattern gene is present in all cats including solid or self cats. Second, the tabby pattern is only revealed when the dominant agouti gene product or protein is present. Third, the recessive non-agouti gene product results in solid coloured cats eg black or blue cats despite the tabby gene being present in their genetic blueprint. Fourth, the non-agouti protein does not work on the dominant 'O' gene pigment colour often resulting in a non-agouti cat with defined tabby markings. These non-agouti red or cream tabbies are often difficult or impossible to distinguish from agouti tabbies and as such fall into an 'inbetween' tabby category that is technically unique to the 'O' gene cats. The issue is further compounded by the tendency of some genetic tabbies to develop a broad solid spine line with age regardless of coat colour.

Historically the agouti gene is named after the fur colour of the agouti mammal that is related to the guinea pig (Nature Com, 2002). The agouti gene protein product creates bands of black and yellow pigmentation on individual hairs. Collectively these coloured bands divulge the nature of the tabby pattern ie ticked, spotted, mackerel or classic. When a black cat has the agouti gene present we see a brown or black tabby[†]. However, when a black male cat has the dominant 'O' gene present we are likely to see a red tabby regardless of whether the agouti or non-agouti gene is there making it very hard to breed solid red or cream cats that are free of tabby markings.

Clear-coated reds and creams

The idea that breeding clear-coated non-agouti reds or creams is almost a genetic impossibility may be difficult to grasp because occasionally we see a red solid Persian or red Burmese that show little or no tabby markings. These magnificent cats are thought to have come about through selective breeding that has enriched for gene modifiers or polygenes that diffuse the tabby pattern. Of the tabby patterns, the ticked tabby is the most amenable to giving rise to a more solid-looking or clear-coated cat. The red Burmese appears as a red self due to being a selectively-bred, non-agouti, ticked tabby that has the added advantage of the Burmese colour lightening gene known as $c^b c^b$ (Vella et al, 1999). However the ticked tabby pattern is unavailable in the Persian gene pool (Brown, 1992). Nonetheless many breeders have been able to diffuse agouti-induced classic or mackerel tabby markings to the point of not being able to easily see banding or tabby pattern by carefully breeding some of the non-red Persian coat colours (Vella et al,

1999). Fine examples of this phenomenon are seen in the chinchilla or golden coat colours (Brown, 1992). Besides being enriched for a special set of polygenes, these colours like the agouti or non-agouti red solid Persian have the advantage of coat length which helps dissipate the classic or mackerel tabby pattern (Vella et al, 1999).

Dense tortoiseshells

Being a sex-linked gene, the 'O' gene colour anomalies continue into the tortoiseshell range of coat colours. Some dense tortoiseshell standards call for an equal mixture of primary colour, red and cream (CCCA Show Standards, 2002). This wording is fundamentally wrong on two accounts. How can a dense coloured cat have cream coat colour when there is no pair of dilute genes in the cat's genetic composition. Understanding why the word cream features in the dense tortie standards is not hard. Obviously when looking at the dense tortoiseshell the 'O' gene coloured patches are often not uniform in colour distribution. Indeed red or lighter shades of red are often present and perhaps for the sake of convenience these paler shades of red have been called cream. Cream is the same pigment as red, only it is spaced further apart by the recessive dilute density gene. Using the word cream in dense tortie standards creates confusion because it is genetically incorrect. This could be simply avoided by using another description for tortoiseshells such as 'Presence of several shades of red are acceptable' (CFA Show Standards, 2000-2001) or 'The colours black and red (pale and dark shades)' (ACF Show Standards, 2002).

The second issue with the black or non-dilute tortoiseshell standards is the requirement of black torties to have an equal balance of black, red and cream. This wording is flawed, not just because of the word cream being there but because it is actually asking for 2/3 of the coat to be generated from the 'O' gene pigment product. A much better description would be for black torties to have an equal balance of black and shades of red coat colour suggesting a 50:50 ratio of black and shades of red.

Sex chromosomes, linkage and X chromosome inactivation

Understanding why the latter description is an improvement requires grappling with genetics and the inheritance of the sex chromosomes. It entails grasping a genetic theory that Dr Mary Lyon, a mouse geneticist from Oxford University was clever enough to work out. What is remarkable about Dr Lyon's discovery is that it applies to all female mammals and as such it is equally relevant to women. Like women and other female mammals, cats are warm blooded, give birth to live young and feed them with milk. Female cats and mammals have two X sex chromosomes whereas males have one X and one Y chromosome. In males and females one of the X chromosomes is always inherited from the mother. The other sex chromosome will be from the father. If the father contributes his Y chromosome then the offspring will be male. If the father contributes his X chromosome then the progeny will be female.

In the tortoiseshell cat the dominant 'O' gene coat colour gene resides on one of the X chromosomes thus enabling a pictorial display of Dr Lyon's discovery that in the female mammals only one X chromosome works in any one cell of the body (Lyon, 1999). Dr Lyon worked this out when thinking about why males do just fine with only one set of X chromosome genes and why females do not have problems with gene dosage by having two X chromosomes. So throughout the bones, organs, muscles, skin and coat of the female cat the genes located on only one X chromosome are producing gene products whereas the genes on the other X chromosome are inactivated early in development. When the X chromosome that has the dominant 'O' gene on it is active then red or phaeomelanin pigment converts black pigment to red revealing red fur and pink skin pigment including pink paw pads and nose leather. On the other hand if the X chromosome that does not have an active 'O' gene is working then no phaeomelanin

pigment is produced instead eumelanin or black pigment colour develops in the skin and coat. The net effect is a female cat with random patches of black and red coat colour.

Male tortoiseshell cats
Occasionally judges are presented with tortoiseshell male cats in different breed sections to judge. There are several genetic mechanisms of how tortoiseshell male cats may come about but in the main the male tortoiseshell has two X chromosomes with one bearing the 'O' gene as well as part or all of the Y chromosome containing the male testis-determining gene ie the male tortoiseshell cat is generally a XXY male (Gould, 1996; Vella et al, 1999). Scientists have shown that it is the expression of the testis-determining gene on the Y chromosome during embryonic maturity that is primarily responsible for the development of the male genetalia before birth in mammals. The absence of the relevant Y chromosome and its specific testis-determining gene or DNA code in the feline embryo results in a kitten with female genitalia. However a female kitten may be born as a male if some of the Y chromosome gene for testis formation is accidentally inherited with the two X chromosomes.

Testis-determining gene resides on the Y chromosome
The proof that the testis-determining gene on the Y chromosome was principally responsible for male sexual characteristics has been demonstrated by a transgenic experiment in the laboratory mouse. Scientists have been able to locate and isolate the testis-determining gene of interest and microinject it into a fertilised female mouse egg. This fertilised egg or embryo was then transplanted into the oviduct or Fallopian tube of a peudopregnant mouse. The recipient or surrogate mouse had been previously mated to a vasectomised male mouse to generate a pseudopregnancy ie the surrogate mouse was able to hormonally sustain a pregnancy but contributed no genetic material to the mouse pups. After transplantation the injected egg spent a few days in the oviduct where it increased in cell number before making its way to the uterus where the embryo implanted into the lining of the uterus. Nineteen days later the recipient mother gave birth to a chromosomally female mouse with male genitalia. Scientists named her 'Randy' and claimed a 'proof of principle' experiment – the putative gene isolated from the Y chromosome was responsible for testis formation in an otherwise female mouse. Like 'Randy' the transgenic mouse, many tortoiseshell male cats are infertile indicating that other genes in the genetic makeup of males besides the testis-determining gene are required for a fully functional fertile male reproductive system (Koopman et al, 1991).

Lessons from the world's first cloned kitten 'Copycat'
The coat colour pattern of the tortoiseshell cat highlights the randomness of the X chromosome inactivation. Some torties are quite black and although these may not fit the show standard they should not be discarded for breeding as they have the same chance of producing red male kittens as their brightly coloured counterparts. The fickleness of the X-inactivation is attributed to events during embryonic development that are not fully understood. A kitten that made worldwide news earlier this year confirmed this. 'Copycat' or 'Cc' is the world's first cloned kitten (Shin et al, 2002). 'Cc' joins a host of other cloned animals such as 'Dolly' the sheep, mice, cattle, goats and pigs. These clones came about by taking a nucleus from a body or cumulus mass cell, transferring it in an enucleated egg and placing the embryonic clone into the oviduct of a surrogate female and allowing pregnancy to develop (Holden, 2002). 'Cc' was born by Caesarean section 66 days after the cloned embryo was transferred to the oviduct of a pseudopregnant queen (Shin et al, 2002).

While 'Cc' is a carbon copy of her tortie and white genetic mother named 'Rainbow', 'Cc' displays some differences in her coat colour (Holden, 2002). 'Cc' has analogous but not identical white markings when compared to her sole genetic parent and into the

bargain ‘Cc’ shows dissimilarities in her tortoiseshell coat colour pattern. These two observations confirmed what scientists and breeders have previously noted about the chance nature of bicolour and tortoiseshell coat colour patterns from one generation to the next. Further studies will be required to identify the precise factors that are responsible for the distribution of the white spotting gene and the random inactivation of the X chromosome during development in the uterus.

Patched blue tortoiseshells versus intermingled blue-creams

While addressing the randomness of X chromosome inactivation it would be amiss not to the address the issue of patching versus intermingling or the much-discussed blue tortoiseshell as compared to the blue-cream. This issue is of particular interest when judging Persians, Exotics and British Shorthairs in Australia because unlike some other governing bodies there are two different descriptions for these coat colours (CCCA Show Standards, 2002). First, the blue tortoiseshell and the blue-cream are both dilute tortoiseshells. Second, they are both examples of what the random inactivation of the X chromosome can do. If the inactivation of X chromosome bearing the ‘O’ gene occurs in well-defined patches of cells then the desired clear patching of the blue tortoiseshell standard will be seen. However, if the inactivation of the X chromosome bearing the ‘O’ gene takes place in small foci of cells that are in close proximity to cells that have an active ‘O’ gene then the desired ‘shot silk’ intermingling of the blue-cream will be observed.

Unfortunately the random nature of the inactivation of the ‘O’ gene produces a number of cats whose appearance is a mixture of tortie patching and intermingling. These ‘inbetween’ cats can cause considerable debate. This debate is further compounded by the other desirable feature of the blue-cream and that is the paleness and pastel tone nature of the blue-cream coat colour (CCCA Show Standards, 2002). Some breeders tend to value the pale coat colour criteria above the intermingling pattern when entering blue-creams and hence pale cats with blue and cream patches are found in the show category for blue-cream. On the other hand, darker coated cats with little patching may be found entered in the blue tortoiseshell category. Both these scenarios leave judges in Australia debating as to whether these cats are better suited to the blue tortoiseshell or blue-cream category. This is a non-issue in some other countries that allow blue tortie patched or intermingled Persians to be shown together in the one category known as blue-cream (CFA Show Standards, 2000-2001).

Tricolours

Further confusion is created in Australia as many breeders register tricolours or blue tortie and whites as blue-cream and whites. Many cat fanciers are aware of the clear demarcation or patching of coloured areas that is observed in dense and dilute tortoiseshells when the white spotting gene is present and yet persist in entering unmistakably patched blue tortie and white cats as blue-cream and white. To add to the confusion around the issue of intermingled blue-creams versus patched blue torties show managers accept entries for the blue-cream and white category despite there being only a listed blue tortie and white show category for judging.

Shell, shaded and tabby cameos

When considering the ‘O’ gene and Persians the ‘inbetween’ colour known as ‘pink’ is also worth discussing. This colour has been used to affectionately describe the coat colour of the shell cameo (Crawford and Crawford, 1993). The definition of cameo is basically a silver cat tipped or shaded with red. The shell variety is tipped with red whereas the shaded cameos have a mantle of red shading. The usage of the word shell in Australia has been strongly linked to cameo with many fanciers overlooking that any tipped silver cat falls into the shell category regardless of hair tip colour. For example the

chinchilla being a black tipped silver cat is also technically a black shell. Likewise a shell cameo is a red chinchilla or red tipped, a shaded cameo is a red shaded and a smoke cameo is a red smoke (Gebhardt et al, 1979). Given this, our current CCCA Group 1 standard needs the umbrella headings of shell cameo and shaded cameo to be amended to shell and shaded, respectively, in order to encompass all the non-red and red colour varieties.

In addition to the shell and shaded cameo categories, there also exists the cameo tabby. These cats have come about by mating red tabbies to shaded silvers or smokes (Krzanowski, 1993). The cameo tabby has caused confusion due to the misconception that the shell cameo as well as other tipped varieties of Persians, has the dominant ticked tabby gene. Instead the Persian tabby gene pool is predominantly classic tabby followed by the gene for the mackerel tabby pattern (Vella et al, 1999) making it quite possible to breed a red silver or cameo tabby.

Apricot – a dominant density modifier gene or a misspelt recessive dilute density gene

The last 'inbetween' that comes to mind when dealing with the 'O' gene cats is the colour apricot which is cream under the influence of a dominant density modifier gene known as $Dm^{\dagger\dagger}$. This dominant modifier gene has also been stated to give a metallic patina to other dilute colours such as blue and lilac, converting them to caramel and taupe respectively (Vella et al, 1999). While there is much phenotypic support for the existence of colour variation in the dilute coat colours, there is considerable debate among Australian cat fanciers as to the genetic basis of these variant colours. The original proposal put forward by Patricia Turner, the foundation breeder of caramels in UK, relies on the inheritance of a single dominant density modifier gene that spontaneously arose in one founder cat in the 1970's (Turner, 1992). While this explanation may turn out to be correct for some phenotypic apricots, caramels or taupes it does not adequately explain the claimed prevalence of these controversial coat colours in many different, independently-bred cat breeds found simultaneously in Australia over the past 3 years.

On the other hand, DNA evidence in mice would suggest that there are numerous alternative gene forms or alleles of the recessive dilute density gene that can also tone or tarnish coat colours (Davis and Justice, 1998). These observations have lead to the hypothesis that there may be a series of alleles for the feline dilute gene similar in principle, to the albino series: cc, c^sc^s, c^bc^b and c^sc^b that alter the black gene in cats to albino, seal point, Burmese brown and natural mink in Tonkinese, respectively (Fowler, 2000). Given that there is little evidence from some Australian cat pedigrees for the direct inheritance of the dominant Dm gene from cats in the UK, a series of mutant alleles or different forms of the dilute gene and/or other related genes that share the same or related pigment pathway, may well explain some of the observed colour variation. Further evidence that different alleles and/or gene modifiers can alter the warmth of coat colours comes from the breeders of Australian Mist cats who have successfully established a relatively new breed of cats with a characteristic rufous mist over their coats. Interestingly, a show colour category for 'caramel' Australian Mists has been recently developed by the Australian Cat Federation (ACF) despite these selectively-bred cats bearing no direct pedigree ties to the foundation caramels described in the UK (Dr Truda Straede, personal communication). If these cats are indeed true genetic caramels then it would suggest that a new spontaneous mutation analogous to the Dm gene has independently occurred in the Australian Mist breed.

At present it would seem reasonable to keep an open mind as to the genetic basis of apricot, caramel and taupe cats from Australia and the UK until scientists undertake DNA sequencing of candidate genes. Meanwhile it would be helpful if visiting experienced

judges from overseas could share their knowledge with Australian judges and fanciers when judging cats of the Dm phenotype, highlighting those cats that have correct apricot, caramel or taupe coat colours. This would enormously help local judges and fanciers in recognising proper Dm-based coat colours as opposed to 'bad blues or lilacs or creams'.

Colour speciality breeding - a bygone era?

It is also worth noting that established cat breeders in Australia have observed odd-coloured blues, lilacs and fawns as well as 'hot' or 'brown paper bag' creams in unrelated breeds for decades. Many breeders have tried to minimise them by reducing the effect of possible allelic variants and/or polygenes by not radically mixing colours when breeding. Also breeders note that the intermating of colours has increased in latter years with specialist colour breeding diminishing. Some fanciers believe that this has been due to decreasing gene pools, the introduction of new coat colours into established breeds and the putting into practice judges' powerful message that 'Type is paramount'.

Time to revisit registration and show standards

In summary, our CCCA show standards give an inconsistent message to breeders and judges of 'O' gene cats. For example, in the dense tortie we ask for red and cream patches to be present, yet we expect their red solid male offspring to show no unevenness of coat colour. It is time that we recognised that many reds and creams with tabby markings have been registered as solids or selfs simply because their parents were registered as such. To overcome this anomaly, overseas governing bodies have allowed registration of red or cream kittens according to phenotype rather than whether there is a registered tabby parent or not. This has some merit given the difficulty in knowing whether a red or cream cat with tabby markings has the agouti or non-agouti gene present. At present our show standards and registration policies for 'O' gene cat needs an overhaul so as to reflect our current knowledge of genetics and minimise confusion. Regardless of whether the CCCA adopts the policy of allowing red or cream tabbies from solid parents to be shown in the tabby section, the pattern of all tabbies ie classic, mackerel, spotted or ticked should be clearly stated on registration papers and in the judges' books. Hopefully, at the end of the day the right changes to registration policies and show standards will result in less controversy when breeding, registering and judging the delightful 'O' gene cats.

Footnotes

† The use of black tabby (CCCA Show Standards, 2002) or ebony tabby (CFA Show Standards, 2000-2001) terminology instead of brown tabby has caused some confusion of late with the Group 2 cats in Australia. Conventional coat colour categories for tabbies are generally described by the colour of the tabby's markings however the brown tabby with its black markings and brown ground colour has in many breeds adopted the colour of its ground colour in the naming process. This has largely come about to distinguish the brown tabby from the silver tabby, which also has black markings (Brown 1992). We need to be mindful that when judging the black or ebony tabby we are actually judging a brown tabby. The simple explanation for this is that the governing body has elected to use the black tabby markings to define its colour category rather than the brown ground colour. Under no circumstance should these black or brown or ebony Oriental tabbies be referred to as seal tabbies because they do not have two copies of the recessive himalayan coat pattern gene known as $c^s c^s$, which acts on the black gene to form seal pointed cats. Neither do they have the Burmese colour gene known as c^b c^b, which modifies black to form Burmese brown, also described in Australian standard as seal brown (CCCA Show Standards, 2002). It should be noted that Burmese brown is described in the USA as sable in recognition of the genotypic difference between c^b c^b and $c^s c^s$ (CFA Show Standards, 2000-2001).

†† The term apricot frequently appears in colourpoint standards as a colour description of red pointed cats that do not have the dominant density modifier gene. If the colour apricot as described by Patricia Turner stands up to DNA scrutiny then another description for the red colourpoints is required.

References

ACF (2002)
Available at: http://www.acf.asn.au/Standards/Persn&Exo_Tby_Tortie.htm

D Brown (1992)
Feline Colour Genetics
Dearinger Enterprises; Utah, USA

CCCA Show Standards (2002)
Co-ordinating Cat Council of Australia
Canberra, Australia

CFA Show Standards (2000-2001)
The Cat Fanciers' Association Inc
New Jersey, USA

S Crawford and D Crawford (November, 1993)
Shady ladies – and gentlemen, too! The beautiful Persians of the shaded division.
Cat Fanciers' Alamanac, November 1993: 7-12
The Cat Fanciers' Association Inc
New Jersey, USA

AP Davis and MJ Justice (1998)
Mouse alleles: if you have seen one, you haven't seen them all.
Trends in Genetics, 14: 438-440

KJ Fowler (2000)
Novel flavours in feline coat colours – lessons from the dilute gene in mice.
NSWCFA Catching Up 2(4): 8-9
Available at: http//www.hotkey.net.au/~f'CC'vic/art12.htm#home

RH Gebhardt, G Pond and I Raleigh (1979)
A standard guide to cat breeds.
Macmillan; London, UK

L Gould (1996)
Cats are not peas. A calico history of genetics.
Copernicus, Springer-Verlag; New York, USA

P Koopman, J Gubbay, N Vivian, P Goodfellow and R Lovell-Badge (1991)
Male development of chromosomally female mice transgenic for Sry.
Nature, 9; 351(6322): 117-21

C Krzanowski (1993)
The Tabby Pattern
Cat Fanciers' Alamanac, June 1993: 9-12
The Cat Fanciers' Association Inc
New Jersey, USA

C Holden (2002)
Cloning. Carbon-copy clone is the real thing.
Science, 295(5559): 1443-4

MF Lyon (1999)
X-chromosome inactivation
Current Biology, 8; 9(7): R235-7

Nature Com (2002)
Available at: http://www.animal-information.com/text/agouti.html

T Shin, D Kraemer, J Pryor, L Liu, J Rugila, L Howe, S Buck, K Murphy, L Lyons and M Westhusin (2002)
A cat cloned by nuclear transplantation.
Nature, 415(6874): 859

P Turner (1992)
The Birth of Caramels.
Courtesy: D Turner, NSWCFA; Sydney, Australia

CM Vella, LM Shelton, JJ McGonagle and TW Stranglen (1999)
Colour Inheritance in Robinson's Genetics for cat breeders and veterinarians.
4th edition, Butterworth Heinemann; Oxford, UK

Acknowledgment

The author thanks Mr Rod Uren and Mrs Marlene Chasemore, Group 1 tutors from Feline Control Council of Victoria and Mrs Lucille Voullaire, Cytogeneticist from Genetic Health Services Victoria, for stimulating discussion. The Coordinating Cat Council of Australia is thanked for the opportunity to present the original draft of this paper at the recent CCCA national judges' conference held in Sydney. Dr Kerry Fowler is Head of Disease Model Unit; a Senior Research Officer with Murdoch Childrens Research Institute, Royal Children's Hospital, Flemington Road, Parkville, Victoria, 3052; an Associate Research Fellow with Department of Paediatrics, University of Melbourne and Public Officer for the Feline Health Research Fund. Email: Janet_Kez@hotmail.com