



The Pigeon Genetics Newsletter

News , Views , & Comments .

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Photos at top of page : King , Anwarul Kabir ; Baldhead Racers , Ismail's Loft ; Fantail , Brian Pogue .

We are on the last leg of winter in Canada , much of the U.S.A. and Europe , while others have no idea what winter is. Regardless , the Pigeons in your lofts are probably busy settling on clutches of much anticipated eggs.

When they hatch , chances are that some will appear almost naked , shiny and pink . That is the theme of this Month's Newsletter " **DILUTION**"Section # (1)
Beginner:



The Gene identified by the Universities in Texas and Utah is Slc45a2 and it is " variation " at that gene that provides us with either Intense or dilute birds . The Intense we know as the "normal" colour.
"Dilution" is a gently washed out version that affects the entire bird's phenotype / appearance .

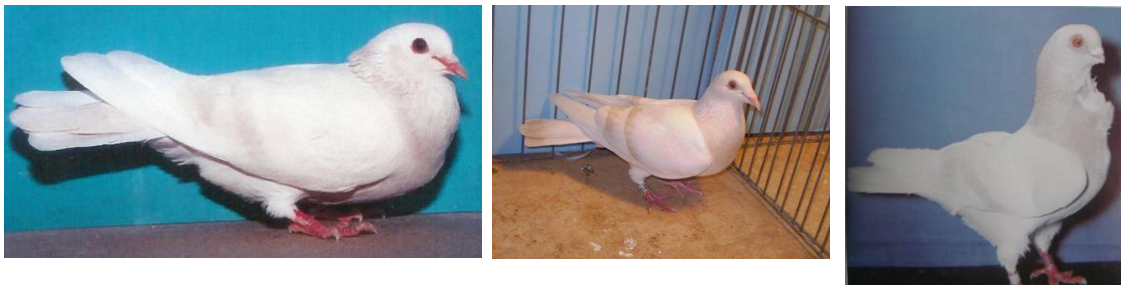
There is also a third phase called **Pale factor** that gives us a tone about 1/4 way between the Intense and dilute phases. These gene mutations are sex-linked . The Intense phase is Dominant over Pale and dilution. Pale is recessive to Intense but dominant over dilution, and dilution is recessive therefore to both pale and Intense.



Pale factor "Gold" in front, & behind Intense bronze "Copper", Riyad Khan photo.

There is a possible fourth phase, which is still undergoing testing . There are some who are convinced that they have enough breeding results to say definitively that it is the extreme phase of dilution that was predicted by Hollander as being possible one day! It has two other names : "Lemon" & "Ecrú". The name Lemon was eventually given by Mr. Barkel after he discovered the mutation in his Blue Checker Racers in South Africa . The name Lemon was moved to describe only Blue Bars in the U.S.A. by Ron Huntley as he gave separate names to the very subtle phenotypes he observed when the gene was applied to the various Patterns and Modifiers . Paul Gibson and other Breeders assigned the name Ecrú for two reasons .. (1) they saw the phenotype as (Muslin tone) and being too similar on all patterns to justify separating them ., and (2) they found evidence that the trait may not be an allele of dilution, but may be very near or overlapping the dilution locus, published Jan. 2010.

Some have suggested that there may actually be two or three different almost identical mutations that are being considered to be one and the same . Others are confusing light dilute ash-yellows , milky dilute ash-yellows, and other phenotypes with the newer mutation.



reduced brown bar milky Indigo , Gibson. Ecrú bred by Gary Boo. milky cream checker , Levi .



No matter what Base colours or modifiers are involved , dilution can play a role if introduced , as it will soften the overall tone . Ash becomes cream ., Blue becomes silver ., and brown becomes tan/ Ochre. The spread forms are more true to the actual pigment : Red becomes Yellow ., Black becomes Dun ., and Chocolate becomes Khaki. Pg.2.

The series Patterns then exhibit a combination of tones : Ash- red barless, barred , checkered , and T-pattern. The dilutes become Cream barless ., cream/ yellow barred, cream/ yellow checker , and cream /yellow T-pattern. For brevity we tend to use shorter terms wherever possible using either just cream ., or just yellow. This is a source of contention because Intense recessive red already has its dilute phase that we call "yellow" , so it can be quite confusing to use "yellow " in both cases, especially for beginners ..

Blue series becomes Silver barless , silver barred ., silver Checker ., & silver T-pattern.

Dilute Brown series becomes : Ochre or Khaki barless ., Ochre/Khaki barred ., Ochre/ Khaki Checker ., & Ochre/ Khaki T-pattern. When you think about what you are saying ., In the case of a "Blue Bar " ., it is a blue series bird with black bars , **thus a blue black bar** , but that is a tad cumbersome , so we just say Blue barred ., thus for the other two base colours , cream barred ., and khaki barred which implies that the bars are naturally darker coarse spread .

Bronze traits have a dilute phase called Sulphur .

Below , Intense recessive red and dilute (yellow) Chinese Owl pigeons from the Facebook Groups .



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# (2) Intermediate : Breeders in England noticed a light silvery coloured pigeon in their lofts back in the early 1800's. Lyell wrote about it in his late 1800's book **Fancy Pigeons** . They noted that all of these were hens , and later found that if these hens were mated back to their sires , more silvers in both genders were produced . Dilution is a recessive gene that is carried hidden once it is introduced. It takes two birds with the genetic trait either present ., or carried hidden in order to produce more. If the hen is Intense and the cock is dilute ., the sons will all be Intense carrying dilution , and all daughters will be dilute. This "Criss-Cross" effect is a sure way to determine the gender of young and should not be confused with another

( Pg. 3. )

genetic term "Crossover" . Breeders sometimes think that a "YELLOW" is needed to make a yellow . Or a Silver is required to make a silver . But in actual fact , you simply need the gene "dilution" from any source mated to the appropriate base colour & modifier to change that trait to its dilute phase , usually taking two generations of young..

If I have a dilute recessive red ( YELLOW) cock ., and a Black hen for example , I can get from them sons carrying dilution , and also carrying recessive red . Their colour will depend upon what base colour the parents are hiding. All of the daughters will be dilute phase of whatever base colour that recessive Yellow masked - ( had hiding) or carries on its matching chromosome. Those daughters could be dilute Ash-red/ creams , dilute blue /silvers ., or dilute browns / Khaki patterns. At least 50% of the young will be Spread factor if the hen is hetero spread factor . The sons mated back to their Dam would be one way to get some Dun females .

Dilution is quite well known and understood by most breeders as far as its use in the day to day breeding practices in our lofts . We still find pockets of fanciers however , who use the gene indiscriminately to end up with weakened birds both in colour and physical attributes. Fanciers working on introducing dilution of a certain colour trait into another Breed , sometimes inbreed extensively , especially while also trying to maintain or improve Breed type at the same time . The result of this has been seen in the Fantail Breed . Generally it is unwise to repeatedly mate dilutes together . The moment you observe the offspring not developing well ., and expressing very washed out colour especially in the (C) Pattern regions., narrow , shallow feathers , and small weak specimens , it is time to make a cross back to Intense and start over .

**Dilute brown** series Khaki bar



**Dilute Ash** series Yellow T-pattern



Dilute ash-red Cream/yellow bar

Dilute Spread blue/ black, Dun and Bronze, Sulphur.



4 above photos : WOE Nat. Show ; Capuchines: ( England ) , Gola Kabra : Mohammad Shoab .



Below Ifi Sonu's Dilute red /yellow, Necklaced Patais an Indian / Pakistanian Breed.



Dilute recessive red / yellow English Trumpeter at Specialty Breeders Show Ontario , taken by Gary Parsons .



Above ash-Yellow Mottle English Trumpeter youngster getting to know my Grand-nephew a few years ago!



Above Post on Face book by Naimal Parves .

Dilute blue ( Silver ) with a **bleached** trait that can be transferred to Blue Intence ( Andreas L. (2000, 2007) , described in Sell's 2014 Book.

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# (3) Advanced .

## Report on Molecular Research of the U of U , by Jith Peter of India .

Before I talk about the research report about Dilute mutation in Columba livia published by the University of Utah, I think it is important to give a brief idea about (1) Some terminologies , (2) Physiology of feather pigmentation in birds, (3) Melanin producing cells ; Melanocyte and its melanin producing organelle, Melanosome, (4) pH is a physiological factor affecting melanogenesis, and (5) function of SLC45A2 enzyme encoded by Slc45a2 gene.

### Some terminologies

Intracellular- Inside the cell.

Extracellular- outside the cell.

Organelle- An organelle is a specialized subunit within a cell that has a specific function. Individual organelles are usually separately enclosed within their own lipid bilayers, Example: Melanosome.

Ortholog- Orthologs are genes in different species that evolved from a common ancestral gene by speciation.

Cytoplasm- Gel like substance enclosed within the cell membrane.

Enzymes- Enzymes are biological catalysts that accelerate or catalyze biochemical reactions.

pH- pH is a measure of the acidity or basicity of an aqueous solution. Solutions with pH less than 7( up to zero) are said to be acidic and solutions with a pH greater than 7(up to 14) are basic or alkaline. Pure water has a pH 7 and is neither acidic nor basic, but normal.

Ions- Ions are charged atoms or molecules

Cation- Positively charged ion, Ex: Hydrogen ion(  $H^+$  ), Sodium ion(  $Na^+$  )

Anion- Negatively charged ion, Ex: Chloride ion (  $Cl^-$  )

Ion transporter- Also called an Ion pump, is a transmembrane protein that moves ions across cells or organelle membranes. Page 7 .

## Physiology of feather pigmentation in Birds

Feather pigmentation is a complex process that occurs primarily in the dermis (skin), beginning with the migration of melanocytes into the dermal pulp of the developing feather germ, where the melanin is produced in the Melanosomes present in the melanocytes and then those Melanosomes are transferred to keratinocytes for deposition into developing feathers. The feather is a complex epidermal organ with hierarchical branches and represents a multi-layered topological transformation of keratinocytes sheets. In keratinocytes, melanin granules are translocated to the upper pole of the nucleus, forming a melanin cap that protects the nuclear DNA from UV rays.

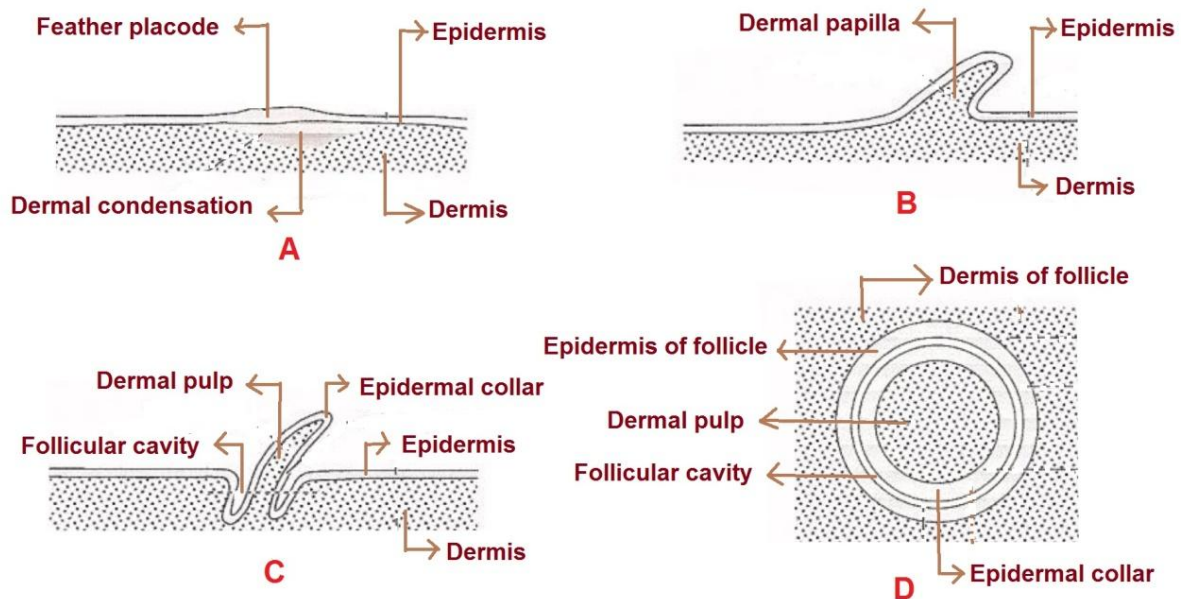
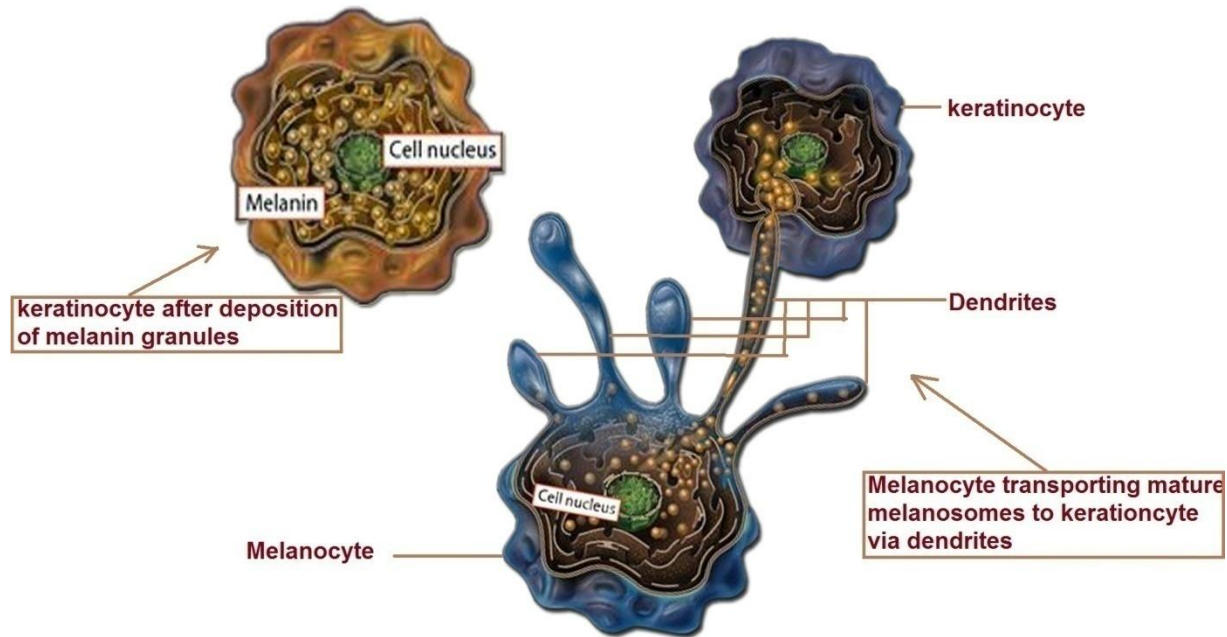


Diagram A,B and C are of different stages of a developing feather germ diagram D is a cross section of developing feather germ.

## Melanocytes and Melanosomes

Melanocytes are specialized cells of the skin that produce the pigment melanin. Melanocytes are branched structures, consisting of a central cell body and numerous branches, or dendrites. Melanosomes are specific melanin producing intracellular organelles produced inside the Melanocytes and they pass through four developmental stages, starting in the middle of the melanocyte cell, and migrating to the outer edges of the cell through the dendrites. Melanocytes can produce eumelanosomes and/or pheomelanosomes.





The Melanosomes structure is different according to the type of melanin produced; pheomelanosomes produce pheomelanin, remain round. Eumelanosomes produce eumelanin and are elliptical. Both types of Melanosomes are typically divided into four maturation stages determined by their structure and the quantity, quality, and arrangement of the melanin produced. Under physiological conditions only the mature Melanosomes are transferred in to keratinocytes.

## Melanosomal pH Controls Rate of Melanogenesis, Eumelanin/Phaeomelanin Ratio and Melanosome Maturation in Melanocytes and Melanoma Cells

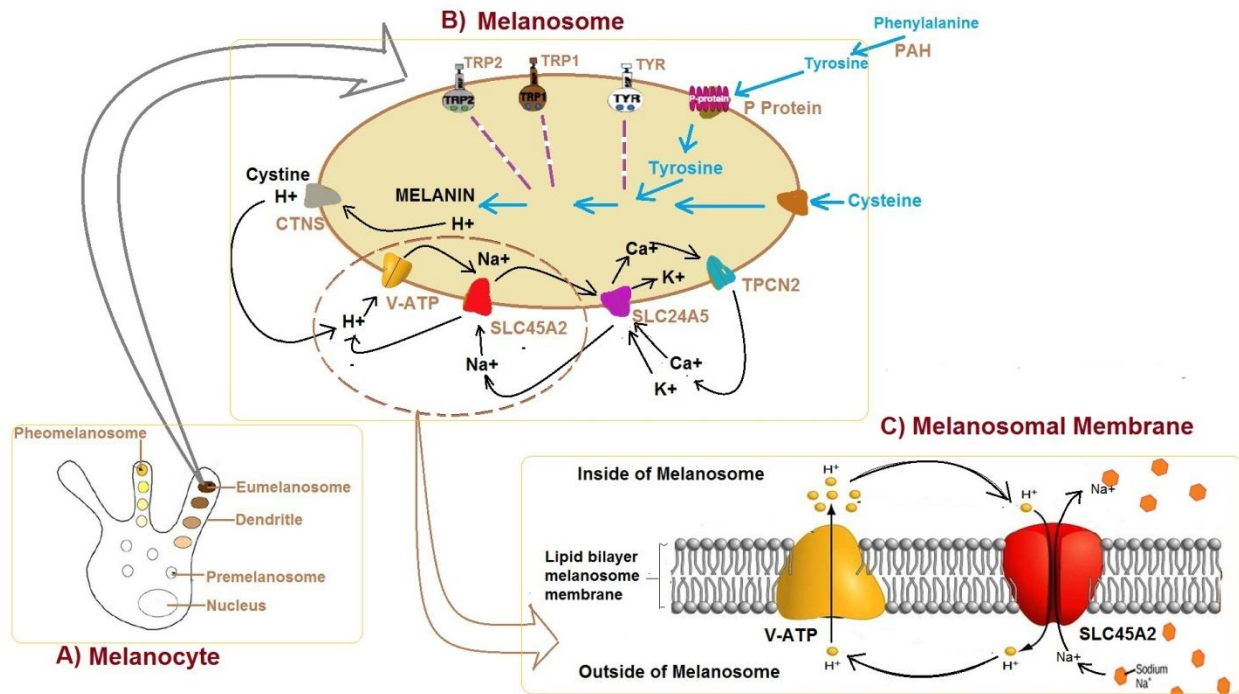
Molecular research on skin colour differences of Caucasian and Negroid human population by University of Bradford and a combined molecular analysis on Zebra fish by Max Planck Institute for Developmental Biology, Tübingen, Germany and Institute of Molecular Life Sciences, University of Zurich, Switzerland revealed that Melanosomal pH Controls Rate of Melanogenesis, Eumelanin/Phaeomelanin Ratio and Melanosome Maturation in Melanocytes and Melanoma Cells.

The differentiation of Melanosomes may require a dynamic range of pH and ionic conditions at different time points. Initially, a low pH may be required for the proper sorting required to form stage I Melanosomes. As the Melanosomes mature, a change in the pH and ionic conditions may be required in order for the optimal activity of tyrosinase to be achieved. The activity of the rate limiting enzyme for controlling the production of the pigment melanin; tyrosinase is known to be dependent on the pH in the Melanosomes, so that varying the internal pH of the vesicle would have a direct effect on enzymatic activity of tyrosinase. The optimal pH of tyrosinase in zebra fish is 7.3, and is maximal at pH 6.8, any further acidic environment would render it almost inactive. In Humans and some other species it is known to vary slightly. However most of this remains unclear and subject of much controversy. Pg. 9.

## Slc45a2 and V-ATPase are regulators of Melanosomal pH homeostasis

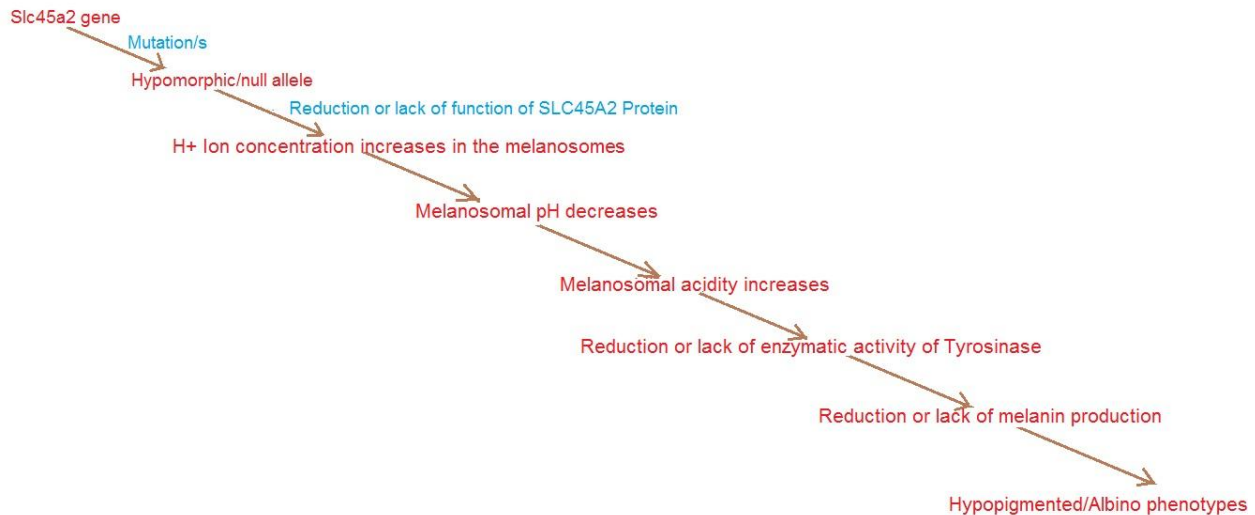
SLC45A2, a transmembrane cation exchanger (Na<sup>+</sup>/H<sup>+</sup> exchanger) localized to intracellular structures consistent with the membranes of melanosomes, is shown to play a role in melanogenesis. Its ortholog is affected in the zebra fish mutant, golden. Slc45a2 Mutations are responsible for various hypopigmented phenotypes and albinos in Humans and many other species like tiger (white tiger), mice (underwhite-like), etc. Studies in zebra fish show evidence for slc45a2, the V-ATP and slc24a5 are involved in melanosomal pH homeostasis required for the normal enzymatic activity of tyrosinase while also playing a role in the exchange of cations(K<sup>+</sup> and Ca<sup>+</sup>) through Slc24a5.

V-ATPase is a highly conserved enzyme found within the membrane of melanosomes and many other organelles. It works as a melanosome acidifier by pumping protons (H<sup>+</sup> ions) across the melanosomal membrane. V-ATPase, thereby raising the pH of the melanosomes. The functional role of SLC45A2 is related to the maintenance of melanosome pH in association with V-ATPase and SLC24A5. The Na<sup>+</sup>/H<sup>+</sup> exchanger SLC45A2 removes H<sup>+</sup> from the melanosome by exchange with Na<sup>+</sup>, which is then cycled back out of the melanosome by the cation exchanger SLC24A5. In this way, SLC45A2 plays a crucial role in the pH and ionic homeostasis within the melanosome. However, there are still arguments about the exact function of SLC45A2.



In the diagram you can see the conversion of Phenylalanine to tyrosine by phenylalanine dehydroxylase (PAH) which takes place in the cytoplasm of the melanocytes (outside of melanosome; where the melanosomes dispersed in melanocytes) and is necessary to maintain the supply of this substrate for melanogenesis to occur continuously. An active uptake of tyrosine by the melanosome is required and is believed to be transported by P-Protein (also known as melanocyte specific transporter or Pink eyed dilution gene). Melanogenesis is initiated by oxidation of tyrosine by tyrosinase and involves other enzymes such as TYRP1 and TYRP2. Ion transport is critical to melanosome function, with TYR activity being pH dependent and its absolute activity is being critical for the rate of melanin production. The coupling of H<sup>+</sup>, Na<sup>+</sup>, K<sup>+</sup>, and Ca<sup>+</sup> transport by the V-ATP complex, with the involvement of SLC45A2, SLC24A5 and TPCN2 in the regulation of this process is shown. Cystine as a negative regulator of melanogenesis is pumped out by CTNS.

Variations in the Slc45a2 gene results in reduction or complete lack of ability of SLC45A2 Protein function, which in turn results in the accumulation H<sup>+</sup> ions in the melanosomes and that results in decrease in the melanosomal pH causing acidic environment in the melanosome. Since tyrosinase activity is dependent on the pH in the melanosomes, varying the internal pH of the vesicle results in reduction or complete lack of enzymatic activity of tyrosinase protein. Tyrosinase is the rate limiting for controlling the production of melanin, reduction in its enzymatic activity results in hypopigmented or albino phenotypes.



Simple diagram showing mutation in the Slc45a2 leads to Hypopigmented or Albino phenotypes.

### Slc45a2 variation underlines dilute phenotypes in Columba livia.

According to the research report published by the University of Utah, slc45a2, is the causative gene for the dilute phenotypes in pigeons. In Humans the gene is present in 5<sup>th</sup> chromosome and around 20 mutations in this gene are responsible for oculocutaneous albinism type 4. To identify candidates for d, they compared the genomes of 5 birds with diluted feather color and 31 birds with non-diluted pigment using VAAST ( a software used to identify gene variations) and mapped in to solute carrier family 45 member 2(Slc45a2) gene which is associated with pigmentation phenotypes in diverse vertebrates, including other birds, but is not orthologous to the dilute locus in the mouse (Myo5a). In pigeons, the d mutation causes a histidine-to-arginine substitution (Substitution is a mutation that exchanges one base for another) (H341R) at a highly conserved intramembrane residue of SLC45A2 resulting in a Hypomorphic allele. A mutation is said to be hypomorphic when the gene activity is reduced but not eliminated/lacked completely.

Additionally, they genotyped 59 diluted birds from 26 breeds and 67 non-diluted birds from 41 breeds and found a strong (but not perfect) association between d genotypes and color intensity under a recessive model. Fourteen birds not homozygous for d had diluted feather color, and one homozygote was reported to have non-diluted color. They suspect the birds which are not homozygous for d with dilute phenotype could be because of other mutations which can lighten plumage like milky, reduced, and faded, and the non dilute plumage of homozygous d could be because of darkeners like dirty, sooty, and smoky. They found only one allele (dilute) and nothing about pale in the report, so might be the birds which are not homozygous for d with dilute phenotype are pales ( another possibility). Page 11.

I have no breeding experience with extreme dilutes (ecru) nor have I seen or read perfect breeding data published by anyone. But from the genetic point of view it is very well possible that extreme dilute and dilute are alleles, in fact some of the OCA type 4 albinos in humans caused by Slc45a2 mutations are rather similar to the extreme dilutes present in pigeons. However, I am still going with the possibilities that it may or may not be in Slc45a2, as there might be other genes which could produce similar mutants so, I believe in all the possibilities.

“Impossible is not a scientific term”- Vanna Bonta .



A very attractive dilute blue series (Silver) bird posted by Ibrahim Geyik on Face book , showing all of the typical features of this mutation The Iris of the eye may be any colour but is usually light , the beak may be dark but is usually horn tip . The flights are usually lighter than the wing bars , tail band , and neck feathers. There is often a bronze crescent on the breast and possibly in the bars and flights basally , this would be sulphur colour..

April 1st., we will look at the 'milky' mutation and all that we know about it . If you have good examples of milky factor on the base pigments alone or in combination with other modifiers , please share ! Until then this is it from the Pigeon Coop ! B.R Editor. Page 12.



Unique pair posted on Facebook by Sheraji Pigeons , dilute Ash-reds . , and bronze .  
Capped white and Capped half -sider with bronze spotted breast and shield trait .  
Shiraji Guldaar



Rollers : Dilution yellow , Intense red, and Pale (Gold ) at the back . Source for this photo and other information on Indian /Pakistanian Breeds provided by Jith Peter , from his files & the Net .