Genetics: A valuable tool for managing and conserving our wildlife

Dr Ben Greyling
Animal Production, Irene
Agricultural Research Council

WRSA
23 March 2018
Genetic menu of the day

- Perspectives on the SA wildlife metapopulation
- Challenges vs. opportunities
- What can we do with Genetics as a tool?
- What have we learnt over three decades?
- in 25 minutes…
- Current trends involving genetics
- Future perspectives
Key message from "Keep calm and let Africa take the lead" conference (March 2018) in Brussels:

“Africa’s wildlife management authorities and the leaders of sustainable use conservation related organizations in southern Africa are successfully managing and conserving their wildlife through sustainable use methods, including hunting”
What role has molecular genetics played in our quest to successfully manage and conserve our wildlife gene pools?
SA’s wildlife metapopulation: Conservancy based vs. commercial ranch based - “..this makes its conservation and management so much more important since the two are invariably interconnected”

- Game ranches: approx. 9000 = 20 million hectares (expectation: around 30 million hectares by 2025)
- National parks: 6 million hectares
- Ranches: approx. 3 times more head of game than in our national parks
Our wildlife also contributes immensely to the GDP through the tourism sector:

- The hunting (trophy and meat) and breeding and game sales sectors: > R20 billion

- Total turnover of the wildlife industry estimated at > R122 billion

**Bottom line**

Both the wildlife ranching and conservation industries have become major and important role players in the economy of South Africa
Central issues relating to populations genetics and …

“What about erosion of our genetic resources and the impact of humans on the evolutionary process and important adaptations necessary for survival and fitness?”

“Our efforts, response time and eventual impact is often hampered and diluted by a disconnect between decision and policy makers, producers, managers, the consumer and scientists”
Some major applications of molecular genetics

• Individual identification – from luxury to necessity - market demand

• Traceability (individual ID), paternity verification

• Pedigree recording (vital for breeding programmes, assessing reproductive success, mating strategies and dominance of individuals)

• Level of relatedness between individuals and populations
Some major applications of DNA technology (cont.)

- Genetic variation and inbreeding
  - Related to fitness, adaptability to change…
  - A proxy for mating patterns, gene flow, dispersal, translocations and reintroductions
  - Effective population size (Ne) - the minimum pop size required to retain levels of genetic diversity
Some major applications of DNA technology (cont.)

• Test for “origin”, migrants (define ESU’s..)

• Species ID and hybridisation between species

• Behavioural ecology, evolutionary biology

• Agricultural biotechnology (including GMO’s, cloning)

• Genomics

  • GWAS – association between genes and traits

  • Quantitative genetics and performance testing
Some major applications of DNA technology (cont.)

• Disease diagnosis and genetic susceptibility to disease

• Phylogeography (structure across landscapes)

• “Bad Boys”: prevalence of deleterious genes and their consequences
Forensics: Stock theft and poaching

2013-2017: > 5000 rhino poached in SA - 95% in provincial parks
- DNA from horn, blood, tissue etc.
- Are our elephant next?
Why do we need Genetics?

- Is the genetic merit or make-up of an animal obvious?
- Can we “see” genetics for all the traits/applications?
- Can we predict how individuals will react/perform?
- Environment has a huge effect on how genes function.

We need to focus mainly on two aspects on the DNA level:

- where individuals (populations) are the same/similar - share the “same” DNA, similar/same phenotypes, e.g. “looks”

– where individuals (populations) have very different DNA – exhibit different phenotypes…

Knowing the above will assist us greatly in decision-making when it comes to breeding and conservation management of our gene pools.
Are we genetically unique or not?
On DNA level – 99.9% similar
The ABC of DNA

- DNA = blueprint of life, the instruction manual for every cell (> 200 types of cells)
- A string of information with 3000 000 000 letters
  ……AACGTCGTGGCAT.. - the sequence DNA = “ID”/code = individual-specific
- All our cells have identical DNA, but cell types differ…
- DNA packaged into chromosomes – contain genes (25 000 in humans, 19,296 in buffalo); 95% = “junk”
- 50/50 concept of inheritance; dominant vs recessive genes
- Creation of variation: 1/10^{-27}
- Environment affects how our genes are expressed = epigenetics
- DNA’s sequence can change (mutate) over time…effect?
Variations in the DNA code

Point mutation (SNP)


deletion

T

insertion

Epigenetics: Same code, but modified to regulate expression

- Modification can be influenced by the environment e.g. alcohol intake, stress, food, chemicals (e.g. smoke) etc.
- Epigenetic modification can be passed on from generation to generation…
- Epigenetic marks are reversible!!
Epigenetics: The Agouti gene
Two animals, same environment, same DNA!

- Unmodified agouti gene (no epigenetics!) = coat is yellow, obese and prone to diabetes and cancer
- Modified agouti gene (epigenetics active), = coat is brown, mouse is thin and has a low disease risk
- Majority of offspring of two phenotypes produce offspring that resemble parents = it means that epigenetic modification is passed on to offspring
- We can feed the fat yellow mice and switch off the agouti gene and her offspring will be thin – reverse epigenetic modification
Epigenetics: Of fear, trauma and growing older..

- Response to fear can be inherited: Cherry blossom aroma vs. electrocution in mice
- When cherry blossom aroma is accompanied by electrocution, offspring also “stressed” when exposed to cherry blossom (NO electrocution)

- Identical twins, the older the more diverse they become – partly because of environment that affects epigenetics

- Children experiencing traumatic childhoods = higher levels of stress hormones = higher risk of developing addictions, mental health disorders etc.
Food and epigenetics

The Queen bee
- Worker bees and queen bee have the same DNA
- Queen much bigger, lives longer..
- Queen fed on better food (royal jelly).

Humans
- **Nutrition** during pregnancy can affect susceptibility to obesity, cardiovascular disease
- Women who *smoke* during pregnancy - their children has higher risk of developing asthma
- Boys smoking at young age – their sons and grandsons one day will have a *shorter life span* than normal
- Studies looking at medicines that can reverse the epigenetic mark that is responsible for development of a cancerous cells due to epigenetic marks that inactivated the gene that products that plays a role in controlling cell division
Examples of the bad effects of mutations/changes
Malignant Hyperthermia (MH): autosomal dominant trait

- MH is a genetic muscle disorder that affects horses, pigs, humans...
- MH is rare, can be life-threatening
- MH due to mutations in the ryanodine receptor gene (RYR1 locus, chromosome 19). Mutation results in massive release of calcium into the cytoplasm - extensive skeletal muscle contraction occurs
- MH mutation may not show any physical signs of the disorder until triggered by exposure to anaesthesia or extreme exercise or stress
Achondroplasia/dwarf syndrome

- **Dominant** genetic disorder
- Affects bone growth
- Mutation in *FGFR3* genes that code for protein involved in making bone from cartilage
- Most people with syndrome have normal parents (80%)
- Dwarf parents have a big chance of passing it on to offspring: 75%
Are all mutations/alleles bad?

**Resistance to HIV**

- CCR5 gene codes for surface protein on cell, HIV recognises this protein to enter the cell. Mutated gene’s product looks different (deletion), HIV cannot enter cell = individuals more resistant to HIV

**Sickle cell anaemia**

Poor oxygen carrying capacity and clotting; more resistant to Malaria (heterozygotes); parasite struggles to attach/infiltrate blood cell

**Double muscling**

*Mutation* in myostatin gene – affects muscle cell growth and differentiation
- more muscle, less fat, but calving difficulties..
4000 genetic diseases/syndromes known…

So you're telling me

That every human has DNA mutations.
GMO’s - changing DNA artificially
Mutations responsible for coat colour: Black Impala and golden wildebeest

- Simple *recessive* mutation

- Can ID carriers / splits for the colour variant – breeding goals?

- Carriers ("splits") do not show colour variant
The colour pattern of the King Cheetah - due to a *recessive* mutation in the *Taqpep* gene
Genetic variation and fitness

We know that:
- High levels of inbreeding and low levels of neutral genetic diversity = reduced adaptive potential, may be more susceptible to disease
- Genetically diverse individuals have superior immune competence

European wild boar

- Increased heterozygosity = increased resistance to BTB
- Certain alleles associated with increased risk to BTB infection
- Certain alleles associated with significant protection
Genetic variation and fitness

- Cheetah considered particularly vulnerable to disease due to low genetic diversity?
- Recent studies suggest that Cheetah have more genetic variation than previously thought
- Other extrinsic factors may also play a role (related to captivity/management)
- Still predicted that cheetah may find it difficult to adapt to change (fitness!) in future…
Genetic variation and fitness

- **Effect of inbreeding depression is complex.**
  - Lions: a mere 5% reduction in male lion dispersal from natal site = substantial increase in the level of inbreeding
  - Lions that have been reintroduced into reserves in South Africa showed tendencies towards inbreeding

- DNA markers for lions available
  - Origin of individuals?
  - Relatedness between individuals - make translocation decisions
Population genetics: What progress have been made in understanding the driving forces behind population dynamics?

Numerous studies have been carried out
- Conservancy-based populations
- Captive individuals
- Ranch populations
The African buffalo as a model species

Populations studied:

- KNP (>37,000 buffalo), HiP (>5,400 buffalo), Addo, St Lucia, SA ranches
- Elsewhere in Africa – numerous conservancies from East to West

Outcome?

- KNP = high levels of genetic variation; HiP = significantly lower
- Extensive gene flow taking place within KNP, no inbreeding
- KNP survived Rinderpest bottleneck much better than predicted…
- No significant differentiation among KNP herds – gene flow!
Significant differentiation/structure between KNP and HiP
- 99% accurate assignment of individuals to population of origin
Both male AND female migration contribute to gene flow

(migration rates are essential for determining the number of translocations needed to keep historical migration patterns intact)

Cape buffalo exhibit significantly lower genetic diversity than its other two cousins, the forest (S. c. nanus) and west African buffalo (S. c. brachyceros)
- Addo and St Lucia populations: Low genetic variation and declining! (1998)

- KNP: genetic variation will be maintained if current population size is maintained

- Little differentiation among Cape buffalo populations throughout Africa, including KNP. KNP share high similarity with haplotypes from east Africa.
More genetics and differentiation
Based on morphology: 4 subspecies
Based on genetics: 2 subspecies

West and Central African populations
- Large genetic differences - manage and conserve lineages separately
- East and southern African populations = very small genetic differences
Is it from the East or from the West…

Notes on assigning an individual to a population of origin

- There must be genetic differences between the populations, thus clear structure
- Availability of reference population
- Markers that can “distinguish” between populations
- Type samples and assign to population of origin as % membership/assignment
- Move away from “purity” % and use association/assignment % to a group?
- Different DNA markers available – can complicate the output: **Talk to your lab!**
Case studies on the prevalence of specific genes and their effects

Its raining men: Sex ratio distortion and the Y-Chromosome

- Environment and body condition “triggers” genes on Y chromosome - so called sex ratio distorters and suppressors that regulate sex ratios

- **SR distorters** - generally on one of the sex-chromosomes - distort sex ratio

- **SR suppressors**: restore sex ratio and fertility

- Both wet and dry seasons characterized by presence of either a distorter gene or suppressor gene that is active

- Sex ratio distortion: poor condition males relatively more fertile in dry season than good condition males
During dry season some males (with specific genotype) mainly produce daughters.

During wet season: some males (with another genotype) mainly produce sons.

This knowledge may be applied in breeding programs.
“Bad boy” genes: The prevalence of deleterious (bad) alleles in our animals: So what?

Background

- Deleterious alleles = negative effect on physical constitution/fitness
  - health, disease resistance (BTB?), body condition, fertility..

- Nature selects against alleles that are bad for you – thus their frequency should decrease over the course of time and become rare

- Too many deleterious alleles = negative effect on population viability

- Recessive (two copies are needed, i.e. the alleles have to be homozygous)
High frequencies of deleterious alleles in buffalo!

- KNP population is enriched with:
  - deleterious alleles with a negative effect on body condition for both sexes
  - sexually-antagonistic alleles with a negative effect on male body condition and a positive effect on female body condition
  - must be a lot of genes (100’s) involved…

- The deleterious and sexually-antagonistic alleles occur genome-wide

- The deleterious alleles are suppressed under certain environmental conditions (e.g. dry years) – epigenetics - thus no effect on body condition in animals born after dry years

- Body condition and health status of one or both parents influence expression of male-deleterious alleles
What happens when deleterious alleles interact with sex-ratio distorter and suppressor genes?

- Positive selection for deleterious alleles = high frequencies of the deleterious alleles - best explained by a relative fitness advantage for males with low body condition

- Male deleterious alleles = low body condition = render animals more sensitive to infection with BTB and stressors such as droughts

- Active sex ratio suppressor genes in males = males have normal fertility, low body condition and carry more deleterious alleles – but have a fitness advantage above males where suppressor is inactive (inactive suppressor = decreased fertility)

**Bottom line:** Low condition males (suppressor active) are more fertile than high condition males (distorter active)!

Thus deleterious alleles result in fitness advantage!
Deleterious alleles spreading northwards to other populations due to the relative fitness advantage for males with low body condition.
Genetics and disease
Alternative methods for BTB management: breeding BTB-resistant buffalo?

- Test and cull methods may not be feasible
- Limited effectiveness of the BCG vaccine in buffalo
- Effective BTB control strategy is complicated by the presence of co-existing/spillover BTB hosts (e.g. lion, baboon, warthog and kudu)
- Disease resistance in breeding (despite relatively low heritability) programmes in cattle have been successfully implemented (brucella/mastitis)
- Genome-wide data: heritability of BTB resistance = 0.21 to 0.23 – selection possible
- A “majority effect” gene(s) could enable us to select for resistance in breeding programmes?
Breeding BTB-resistant buffalo an option?

We know that:

- Genetic markers linked to BTB: interferon gamma gene
- Genetic markers exist that are linked to body condition
- Genetic markers recently discovered that are linked to BTB status (SNP’s)
- High body condition lowers susceptibility to diseases
- Marker-assisted selection (and breeding) for low levels of deleterious alleles is expected to result in animals with relatively high body condition that have increased resistance against BTB

✓ Future option to identify animals that are predisposed to disease susceptibility
Wildlife ranching: From perceptions to facts

- > 20 million ha in SA under ranching; (60 000 buffalo?)

- Generally small populations

- Controlled breeding, selection and gene flow

- Fragmented populations and “lines”

- Selective breeding for superior genetics

- Need to strike a balance between traits of economic and biological efficiency
Potential implications of ranching with wild animals

Human intervention:

- Gene flow/mating mediated through management practices
  - Reduction in genetic variation (inbreeding)?
  - Loss of adaptability/fitness (disease resistance, reproduction, growth etc.)?
  - Increase in recessive/deleterious alleles?
  - Loss of alleles and gene fixation?
- Small populations are prone to genetic drift
  - can we sustain variation?
- Admixture – potential outbreeding depression
Case Study: Population genetics of the ranch buffalo metapopulation

And then?
Formulate and recommend management policies to improve genetic diversity in herds and introduce mitigating efforts if, when and where necessary

- Dataset comprised of about 4000 buffalo genotypes, 26 ranches, 34 microsatellite markers
What did we find?
Most ranches exhibited heterozygosity's (genetic variation) comparable to that of KNP, no inbreeding due to incestuous matings and relatively little movement of buffalo between ranches.
Genetic differences among buffalo populations/herds

- Males and females from the same ranch have different origins, also relatively unrelated – an indication of outbreeding
- Female buffalo also seem to be derived from a more diverse stock than male buffalo
- Males are more closely related amongst each other than females are
Genetic differences among buffalo populations/herds

- Relatively large genetic differences among ranches – relatively little gene flow between ranches?
Genetic differences among buffalo populations

- Buffalo from ranches most closely related to Kruger buffalo
- Fewer deleterious alleles than in KNP
• Low genetic diversity (< HiP) in only 4 out of 22 ranches - could be attributed to the small number of individuals (small herds) and **not** due to inbreeding.
Some ranches exhibited more genetic variation than KNP and HiP together – derived from more source populations?
Take home message from this case study?

- Our buffalo on ranches are in good genetic health
- It confirms the responsible and sustainable management practices on ranches and also highlights the value of molecular genetics as a valuable tool for this prolific industry

“Wildlife ranching and the private sector have generally had a positive effect on this species as it has been widely reintroduced onto private properties within its natural distribution and introduced on those outside of its distribution. Private landowners have also bred numerous disease-free herds that can be reintroduced into protected areas. There is an increasing trend to breed buffalo under intensive conditions due to the high value and demand attached to the species”"
Recent developments in wildlife ranching: Performance recording

Goal: To improve biological and economic efficiency of animals

✓ Gather baseline info on traits

- Fertility, reproductive performance, scrotal circumference, horn colour variants, functional efficiency, carcass quality etc.
- Heritability of traits: some traits are easy to record (e.g. SC) and also moderate to highly heritable (0.46 - 0.75) - can be selected for
- Some traits are also correlated with others, e.g. SC correlated with female reproductive traits (age at puberty and at first calving)
- Qualitative traits e.g. coat colour – one or a few major genes involved – not affected by environment
- Quantitative traits – many genes involved, all with different and varying effects on a trait – e.g. reproduction, - affected by environment
- Pedigree verification!
Recent developments in wildlife ranching: Performance recording cont.

✓ Develop/make use of benchmark/proxy as a selection tool?
- Selection for single-gene traits relatively simple – progress possible in a relatively short period of time
- Multiple-gene traits are more complex, progress take longer

✓ Ultimate aim: predict the genetic merit of an animal as a parent (for one or more traits) making use of breeding value technology
Use data to enable selection in line with breeding goals

✓ Monitor impact and progress – is there a return on investment?

**Beef cattle as a model: 1970-2014: 32% return on investment**
On the horizon…Genomics

Overall objective: To fast track genetic improvement

- Based upon the association of a DNA sequence and a trait

**Principle**
Combine performance/phenotypic data with pedigree data and DNA sequence data – predict genetic merit of an animal based on genomic breeding values…

**Benefits:**
- select more accurately
- select at an earlier stage (shorten generation interval)
- get a handle on traits that are difficult or expensive to measure

**Cattle as a model species: significant returns on investment into Genomics experienced worldwide**
Future and take-home-message

✓ Genetics as a science has contributed immensely to our ability to make better conservation/management decisions for our wildlife industry

✓ We are in a position to respond more timeously and effectively if needed by implementing available technologies

✓ New opportunities arising as a result of technological developments:
  ✓ full genome sequences becoming available (and cheap!), gene markers developed, GWAS in progress, fast tracking of breeding and selection, diagnostics, pop. genetics etc.
Future and take-home-message

What are some of the prerequisites for reaching a “fairy-tale” position?

✓ Availability of baseline information
✓ Access to and availability of samples, sample repositories, genetic markers
✓ Generation of data and databases
✓ Computation technologies
✓ A healthy relationship with your genetics laboratory and knowing what your service provider can offer!

“But for most managers and conservation biologists, the methods and results of molecular genetic surveys are a mystery…”

“We need to inform end users in disseminating conservation technologies”

✓ Buy-in and cooperation among all stakeholders of the wildlife fraternity
Thank you for your attention
Any questions?

“Try to make sense of what you see and wonder about what makes the universe exist....” - Stephen Hawking