Fowl play

A long-term study is revealing surprising insights into male sexuality and fertility. **Tom Pizzari** explains how the humble chicken is revolutionising the study of reproductive biology.

What makes some men more fertile than others? This is a topical question as we face progressively more severe fertility problems, often due to male factors. Of course, why some males leave more offspring than others is also a central question if one is interested in the way populations of plants and animals grow and evolve. Because male reproductive success is so variable, selection based on the competitive ability of males to acquire partners and fertilise their eggs is one of the strongest known evolutionary forces. A force that drives the evolution of some of the most extravagant traits: from the giant sperm of some fruitflies – 20 times longer than the male himself – to bird song, from the peacock's train to… well, sports cars. Understanding the evolution of male fertility is why our group studies male sexual behaviour. And this is where roosters and hens come into the story.

Petra Wagner/Alamy

While male sexual promiscuity in animal societies has always been obvious and culturally accepted, a widely held traditional view saw females of most species as strictly monogamous – copulating with only one male. The advent of molecular tools to assign paternity some 20 years ago revealed that this view was merely Victorian wishful thinking. Two decades on, we now know that in most species both males and females are sexually promiscuous. In fact, females often actively seek to mate with different males. This means that not only must males compete with each other to mate with a female, but because she may also mate with other males, their ejaculates will continue to compete to fertilise her eggs. In other words, whenever females obtain sperm from multiple males, male reproductive success is determined by social mechanisms that enable a male to access and inseminate a female, and after insemination, by the interactions between competing ejaculates within the female reproductive tract. While we are still discovering the rules of engagement between competing ejaculates, we do know that across a range of species two general characteristics make an ejaculate competitive. First, the number of sperm inseminated by a male into a female relative to the number of sperm that competitors inseminate into the same female. Second, the swimming performance of inseminated sperm.

In social species, fathering offspring is often a matter of being socially dominant to monopolise females and inseminating competitive ejaculates. The fowl is a typical example of this.

The red junglefowl is the wild ancestor of our modern day domestic chicken. Red junglefowl populations in south-east Asia live in socially structured groups comprising several males and several females where sexual promiscuity is rife. Socially dominant males attempt to monopolise groups of females and guard them from the advances of socially subordinate males. The subordinates steal copulations whenever the dominant male isn't looking.

Fowl provide a terrific opportunity to inspect closely the mechanisms that determine variation in male reproductive success both before and after insemination. Our group at the Edward Grey Institute of Ornithology, within the Department of Zoology of the University of Oxford, studies the sexual behaviour of red junglefowl under natural conditions.

Evolutionary biologist Hanne Løvlie collects natural ejaculates from male fowl in an experimental study of a feral fowl population.

We borrow techniques from the poultry industry to evaluate sperm quality and use domestic fowl lines, artificially selected for different traits, to work out the evolution of reproductive traits. We also use powerful genomic tools afforded by the recent sequencing of the chicken genome to identify the genes associated with male fertility. The combination of these approaches is building a multi-dimensional picture of male sexuality and fertility which is turning reproductive biology on its head.

An enduring challenge in the study of male fertility is to understand the relationship between male social status and ejaculate quality. In 2002 we started a collaboration with reproductive physiologist David Froman at Oregon State University, who had developed an analysis of new sperm quality that is a powerful predictor of the fertility of an ejaculate. We found that males that produced sperm of low quality were on average dominant over males producing high-quality sperm. These results suggested that perhaps there was a trade-off between what a male can invest in social status and what he can invest in sperm quality.

We decided to head back to Oregon to investigate this idea further and, as is often the case with follow-up studies, we found the situation to be more complex than initially envisaged. We wanted to establish the relationship between status and sperm quality by asking, 'What happens to the sperm quality of a male when his social status changes?' This was a daring question because reproductive biologists do not normally entertain the idea that the sperm quality of a male may change due to external social factors.

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To answer this question we found pairs of males where one was dominant over the other, and then swapped males across pairs to match dominants with dominants and subdominants with subdominants. Because there can only be one dominant male in a pair, this swapping forced one of the males in each of the new pairs to change status. Our experiment revealed that the sperm quality of a male, far from remaining constant, increased or decreased over time, and crucially for us, some of these fluctuations were associated with social switches.

We found sperm quality dropped sharply in males that had become dominant. So social dominance may come at the expense of sperm quality. This suggests that males can adopt one of two alternative strategies. A male may become socially dominant, mate repeatedly with several hens and reduce the risk that his ejaculates will compete with the ejaculates of other males. Or the male can become subdominant, copulate less but inseminate more competitive ejaculates.

While these results fitted nicely with the idea of a status versus sperm quality trade-off, something wasn't quite right. Some of these changes were happening surprisingly fast.

It takes about two weeks for young sperm cells to develop into functional cells ready for ejaculation. If status switches influence sperm quality by changing the way new sperm are produced, one would expect changes in sperm quality no earlier than two weeks after the switch. But while some changes in sperm quality occurred two weeks later, we noticed others occurring within three days of a switch. These changes could not be explained by new sperm production. Instead something appears to happen to sperm already manufactured that influences fertilising efficiency. The most parsimonious explanation for these ultra-rapid fluctuations in sperm quality is that seminal plasma, the liquid in which sperm mature in the male reproductive tract and the liquid part of the ejaculate, may influence sperm performance. Natural physiological changes, such as possible fluctuations in the levels of glutamate or calcium caused by the social challenge of establishing dominance over another male, may change the biochemical composition of seminal plasma which may in turn influence the performance of fully-developed sperm.

A good experiment is said to raise more questions than it answers. In this respect our study was a success. Much of our current research is turning its focus on the role that seminal plasma plays in the battle for fertilisation. ❖

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