

More On DNA and Dairy

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NEWARK, Del.—Recently I wrote about DNA as the latest and most powerful tool for progress in modern animal science and animal production.

I reported on the reliable new method for finding out whether a calf is a red-color barrier. Now, there is a second DNA-based procedure in the very important area of sex determination.

For years I have wondered about the question of sex determination. In our University of Delaware herd and in others I have observed a preponderance of male calves when older semen ampules from famous bulls were used.

Often I received bull semen donations from Holstein or Guernsey breeders at their dispersal sale.

The semen typically has been stored for a time and is often from older bulls that have reached fame as superior performers by the majority of daughters they've pro-

duced, and a performance reputation takes time to establish.

Although I had discussed this older semen issue with bull stud managers, I never succeeded in a research project to collect appropriate data for reliable analysis. I am still convinced there is something to this conjecture.

We know that there are two kinds of spermatozoa: those that determine female sex and those that determine male sex in offspring.

And the physical difference between the two is that the male sex-determining spermatozoa have only the small Y chromosome, while the female-determining spermatozoa have the big X chromosome.

If there is a difference in size, that must also mean a difference in weight or gravity or migration during storage.

When semen is stored, some spermatozoa could settle out because of greater weight. If more of the big-chromosome spermato-

zoa settle out than the small-chromosome ones, this could explain why more of the small male chromosome spermatozoa are left in "solution," which, when used on insemination, would produce mostly male calves.

This is a plausible answer to my puzzle of too many bull calves from older semen—at least more than a 50 percent chance.

A reinforcement of this theory came from botany.

I knew that when pollen grains are added to a blooming flower's stamen, they migrate down inside toward the egg cells of the flower for fertilization.

But while they move downward, some pollen grains travel faster than others. It's the male pollen with the smaller Y chromosomes that migrates faster.

This has been proven by cutting the channel in half at a certain time interval and physically preventing the slower female pollen from reaching the eggs, thus producing a different sex ratio in the

seed.

Does that not also sound like the case of semen from older bulls that has been in storage for some time?

Well, over the years, many scientists have tried to find methods to separate Y from X spermatozoa.

Electric currents and vinegar douches have often been successful, but the semen died in the process, much like the proverbial surgery that was successful except that the patient died.

Now along comes DNA, the new magic.

Of course, for some time, we have been able to see under the microscope whether a fertilized egg is male or female. The challenge has been to predetermine, in a practical way, which egg is going to be male or female by sorting out the respective semen.

Remember, it is only the spermatozoa that make the difference; eggs are always only female, because females (cows) are

homozygous for the X chromosome, while males (bulls) are always heterozygous for X and Y chromosomes, producing them half and half.

Mating with the homozygous female X chromosomes follows the third law of the discoverer of genetics, Gregor Mendel, which states that the offspring numbers between a homozygous and a heterozygous parent are always in a 50:50 ratio; i.e., half of one parent's type, homozygous, and half of the other parent's type, heterozygous.

Unlike the situation with black or red color in Holsteins (where black is dominant over red and red is not visible in the animal's appearance unless it is homozygous for red), in sex there is no dominance.

A heterozygous animal for sex is visibly male and there is no homozygous YY animal, only heterozygous XY and homozygous XX.

The story of the new DNA test for finding and separating the male determining Y spermatozoa comes from research at the USDA Experiment Station in Beltsville, Md., by Dr. Larry Johnson (Dairy Profit Report, University of Wisconsin-Madison, vol. 7(2):1-2).

Johnson determined that X chromosomes have about 4 percent more DNA than the smaller Y chromosome; he used this fact to separate spermatozoa.

Spermatozoa are treated with a bluish fluorescent dye, and because X spermatozoa contain more DNA than Y sperm, the X sperm absorb more dye.

This then is measured by an optical detector in a so-called flow cytometer, which can analyze and sort microscopic cells.

Also, there, the more luminous X sperm are earmarked with a positive charge, the less luminous Y sperm with a negative charge.

Invisible to the naked eye, the dye-charged sperm flow single file past deflecting plates, separating the positive and negative sperm into two containers.

When cow ova are fertilized in vitro by the separated sperm, the success rate of obtaining male or female calves has been 90 percent.

With a new procedure of harvesting ova from cows' ovaries directly and repeatedly, called ultrasound-guided follicular aspiration, the use of the separated sperm is ready for practical wide use, because much less sperm are needed.

The cytometer separation of sperm is actually not fast, requiring about 40 hours to separate 20 million sperm needed for one artificial insemination.

For the in vitro insemination, only a few thousand sperm are needed, thus 200 to more ova can be fertilized from separated sperm sorted in one hour. In a study with a British company in 1992, predetermined 40 bull calves were produced from 50 births.

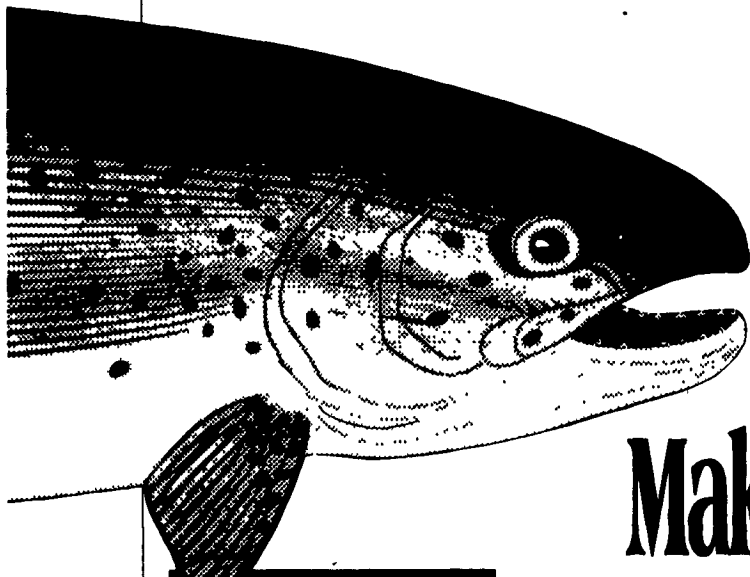
So AI (artificial insemination), the great dairy success story, and ET (embryo transfer), the more recent extension of that success story, now have the culmination of progress potential for breeding only one sex from the top cows in the herd with the top bulls in the world.

What a difference from grandfather's cow farm! And what a difference in profit potential!

What next? In terms of world ecology and environment, maybe

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Pennsylvania Streams and Lakes



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LOST STREAM MAP FOUND AND RESTORED

THE STREAM MAP OF PENNSYLVANIA was completed in 1965 after a thirty-year effort by Howard Higbee, a former Penn State Professor.

The map is also known as the LOST STREAM MAP to some anglers.

Professor Higbee succeeded in creating a map of the highest detail possible...a map that shows every stream and lake. He painstakingly plotted by hand, the location of 45,000 miles of streams onto a 3 x 5 foot map.

The map sold extremely well -- until it was lost several years later. Incredibly, the printer entrusted with the original drawing and printing plates, declared bankruptcy, then carelessly hauled Higbee's 30 years of work to the landfill.

The few remaining dog-eared copies became a prized fisherman's possession. Professor Higbee was offered \$400 for one of his last maps. And state agencies were forced to keep their copies under lock and key.

The experts had always told Professor Higbee that reprints were impossible, because the maps were printed in non-photographic blue.

Then, in 1991, at the age of 91, Howard Higbee's dream came true. Computers made it possible to reprint the map. Holding an updated map, Howard said, "I never thought I'd live to see this day."