

First in vitro twin calves born in Pa.

BY TRISH WILLIAMS
QUARRYVILLE — The first twin calves ever to be born from in vitro fertilization were very appropriately delivered on Labor Day, but without any labor to their true mother. Penn's Pride and Quintus, as the twin bull calves were christened, were conceived in tissue culture glassware, and carried to full term by a surrogate mother.

Born in southern Lancaster County at Pennstar Embryo Transfer Clinic, the twins are the culmination of years of in vitro fertilization and embryo transfer research at the University of Pennsylvania, Veterinary School.

Penn's Pride weighed in at 65 pounds at birth and Quintus at 45 pounds, according to Dr. Troop the attending veterinarian. They were born nine days early, said Troop, but that is quite normal for twins.

Another set of twin calves, also the result of in vitro fertilization research at the University of Pennsylvania were born dead the week before. The calves appeared normal and went to full term but had apparently died from calving difficulties, reported research specialist Karen Bennet. Thus Quintus earned his name as the fifth full term calf to be born from the in vitro fertilization process.

Project leader for the in vitro study, Dr. Benjamin Brackett, referring to the study said, "It is a collaborative effort between the University of Pennsylvania, Atlantic Breeders Cooperative, and Dr. Troop."

Working on a grant from the National Institute of Health, with support and fresh semen supplied by Atlantic Breeders, and donor and recipient cows supplied by Dr. Troop, the team has broken new barriers in the realm of animal reproduction. Dr. Brackett a reproduction specialist and PhD biochemist, won world acclaim when, Virgil, the first calf to be born from an in vitro fertilization was born on June 9, 1981.

Brackett has been involved in in vitro fertilization research since graduation from the University of Georgia, Veterinary School in 1962. After completing a PhD in biochemistry, he joined the Department of Obstetrics and Gynecology at the University of

Pennsylvania, and was given a joint appointment in the Veterinary School. In 1974 he was promoted to professor of animal reproduction in the Veterinary School, and thereby switched his major and minor appointments.

Dr. Brackett presently serves on the Board of Governors of the International Embryo Transfer Society. Several years ago Brackett was called on by the United States Congress to review the field of animal reproduction and the associated new technologies.

"Congress was especially interested in predictions for the future," explained Brackett. "So we convened a small group of experts in artificial insemination, embryo transfer, and even cloning and genetic engineering. We then published a book two years ago entitled New Technologies in Animal Breeding."

Brackett edited the book, and helped to author it as well.

"Technology has evolved rapidly," said Brackett. "We could probably add a chapter or two to the book just from research in the last two years since the book was published."

Speaking more specifically about his own research, Brackett explains "Most of our work has been carried out in rabbits, with later extension to the cow. In vitro fertilization is now feasible in cows, we proved that with the birth of Virgil in 1980."

"The real challenge has been to treat the sperm outside the female reproductive tract," Brackett claims. "The sperm must be conditioned or capacitated to be able to fertilize the egg."

Much of Brackett's research has focused on in vitro sperm capacitation. "We learned how to do this through a series of experiments. We learned that we could add a high salt medium to glassware containing the sperm to remove proteins from the sperm surface, that would enable the sperm to penetrate the egg and fertilize it."

The egg is recovered surgically from the donor cow. In a warm room (38 degrees Centigrade), the egg is inseminated by combining it with fresh 'raw' semen in tissue culture glassware. Raw semen



The first live twin calves born from in vitro fertilization, were born on Labor Day in southern Lancaster County. The twin bulls, Quintus, left, and Penn's Pride, were conceived in tissue culture glassware at New Bolton Center, the University of Pennsylvania, Veterinary School. Pictured with the twins are Dr. Carl Troop and the surrogate mother.

meaning that it has not been treated with preservatives or extenders. The fertilized eggs are cultured in the warm room for a couple of days in the warm room, by that time they have reached the 4 cell stage and are surgically transferred into the oviduct of a recipient cow.

"This is a surgical procedure the way we do it now," Brackett explains. "We hope to develop it, to make the recovery of eggs easier, by recovering by means of laparoscopy. And if we can culture the fertilized eggs a little longer in the warm room, we should be able to transfer them into the uterine horn through the cervix, non-surgically."

What are the advantages of in vitro fertilization over in vivo fertilization?

—The first advantage Brackett points to is to overcome infertility. "For example if eggs have been collected repeatedly from a cow, then the man-made damage might preclude sperm reaching the egg or undergoing capacitation, properly becoming prepared for fertilization. Yet we may still be able to get eggs that are normal from the cow that we can transfer into another cow. There are already a number of valuable cows that are candidates for this procedure. I think within a year this procedure will be in place as far as the initial efforts to overcome infertility in these animals."

—To maximize the utilization of semen from a valuable bull and eggs from a valuable cow. For example there are a number of very valuable bulls that are dead, and only a limited number of ampules of their frozen semen remains. It is desirable to have as many calves as possible from these valuable animals. So by bringing a few sperm cells from the frozen semen into close contact with a number of eggs from valuable cows, it should be possible to get more offspring than might be obtained with just an insemination of the ampule into one cow, where you expect to get only one calf.

—To rapidly assess the fertilizing ability of sperm. This would greatly expedite young sire proving.

—In vitro fertilization used in concert with genetic engineering can very rapidly improve livestock. Such genetic engineering techniques as nuclear transfer, or direct injection of desirable genes into the pronuclear eggs, would be easier with a large number of synchronously developing embryos.



Brackett is pictured here with Virgil, the world's first in vitro calf. Virgil was born in June 1981, and still remains at New Bolton Center, the University of Pennsylvania's large animal research facility.

Reflecting back on his research and many accomplishments in animal reproduction, Brackett relates, "The most exhilarating thing is to look at human in vitro fertilization as an accepted clinical procedure today. This has evolved

from the sort of research that we have carried out through the years, to improve the procedure, to define the conditions, develop the medium, that enable fertilization to occur in vitro."

\$90,000 semen theft

WESTMINSTER, Md — An estimated \$90,000 of Holstein bull semen was reported stolen from Coldsprings Farm Sept. 8, by Marlin K. Hoff, owner of the farm and the stolen semen.

Hoff, who serves on the board of directors for Sire Power, Inc., said he believed the theft had to have occurred between 9 to 12 p.m. Thursday night while he was driving to Sire Power headquarters in Tunkhannock, Pa. to attend a board meeting the following day.

"The semen could be accounted for at 9 p.m.," said the Carroll County dairyman, "But at 12 p.m. our hired man noticed the tank was missing. He thought I must have taken the tank with me to Tunkhannock, it was not until the following day that the theft was reported to the State Police."

Two liquid nitrogen canisters were taken from a storage shed. Hoff said the two canisters, one 5 gallon and one 2.5 gallon, contained only Holstein semen. Most of the semen was from very

popular sires including, Elevation, Tony, Chairman, Spirit, and Bell. Approximately 95 percent of the semen was in French straws, the other five percent was in ampules, reported Hoff.

Hoff was not carrying any insurance on the semen, and will sustain a total loss on the stolen semen.

Because farmers and "semen jockeys" buy and sell semen among themselves, and there are no unique identifying marks on the straws or ampules, Hoff gives very little hope of being able to track down and recover the semen.

In a phone conversation Hoff stressed, "If a farmer does not want to buy stolen semen, they should buy only from certified representatives of bull studs."

Maryland State Police are urging anyone who suspects they are being sold stolen semen, or who has any information relating to the stolen semen to contact them at the Westminster Police Barracks, phone (301) 848-3111.



Dr. Benjamin Brackett headed up the research team at the University of Pennsylvania that is responsible for the birth of the twins. Most of Brackett's research has been carried out in rabbits, and then extended to the cow.

