



# Ovum Pick Up and In Vitro Production in the bovine after use in several generations: A 2005 status

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## Abstract

The first In Vitro Produced (IVP) calf was born in 1981 and the non-surgical Ovum Pick Up (OPU) technique for the bovine was adapted from the human in 1987. Since then, considerable research has been aimed at improving both technologies in the bovine.

Both OPU and IVP can now be seen as mature technologies. It can be estimated that more than 200,000 IVP calves have been born world wide to date, and when the two technologies are combined they are capable of producing over 50 calves per donor cow per year, albeit with a large variation between donors.

Not many new breakthroughs are expected for OPU. For IVP however, automation and miniaturization as well as a greater understanding of the embryo through the application of gene based technologies such as micro-arrays, may provide an in vitro environment that is more in vivo-like than traditional micro drop/well systems. This improved environment should result in higher embryo developmental rates as well as improved quality and welfare of subsequent offspring.

The application of OPU/IVP has progressed from treating infertile high genetic multiple ovulation and embryo transfer (MOET) cows in commercial situations to enhancing breeding scheme designs. With the bovine genome being rapidly sequenced and bovine genes for traits of economic interest becoming available in the coming years, OPU/IVP will prove invaluable in rapidly multiplying rare genes or Quantitative Trait Loci (QTL) of high value. In due course, it is anticipated that Marker Assisted Selection or Gene Assisted Selection (MAS/GAS) schemes will be more widely implemented. In addition, OPU, and particularly IVP, provide the basis for more advanced technologies such as cloning and transgenics.

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This paper is dedicated to celebrate and recognize the significant contributions made by Theo Kruip (1939–2003) to the wide area of bovine OPU and IVP.

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## 1. Introduction

After its introduction in the 1950s, Artificial Insemination (AI) became more and more popular in the 1960–1970s, once the advantages of the technology became clear. Bull stations and herd-based systems for genetic evaluation were established. Prevention of venereal diseases and more importantly, the possibility to generate multiple offspring per bull in a short time frame allowed genetic evaluation of bulls through the performance of their daughters and the first breeding schemes were developed. Once genetically superior bulls were identified and selected, AI contributed once more to the rapid dissemination of the superior genes of bulls into the national and later also international cow populations. In the early 1970s, superovulation or multiple ovulation and embryo transfer (MOET) was developed, which involves administration of hormones to donor cows to induce multiple rather than a natural single ovulation. Ovulated oocytes are fertilized after routine AI and zygotes are allowed to develop into embryos and migrate through the oviduct into the uterus. At Day 7 after insemination, the embryos are in the uterine horns and can be flushed from the uterus and recovered from the flushing fluid. Initially all embryo recoveries and transfers were performed surgically although by the late 1970s non-surgical techniques for both embryo collection and transfer were developed and have become mainstream procedures. Some advantages of MOET include higher selection intensity on the female side in breeding schemes (fewer donor cows could produce more offspring or bulls per time unit, cow to bull selection pathway) and increased selection accuracy (more full and half sib information). However, MOET results still vary highly among donor cows and MOET is not effective in generating multiple offspring within a limited time span for all donor cows.

Following the birth in 1978 of Louise Brown, the first IVF baby [1], research efforts into developing similar techniques for the bovine increased. Initially, mature human oocytes were recovered percutaneously through the abdomen under local anesthesia in the late 1970s to early 1980s [2]. Similarly, mature oocytes were surgically recovered from the ovaries or oviducts of hormone-stimulated cows and subsequently were fertilized and cultured in vitro before transfer and resulted in 1981 in the birth of Virgil, the first IVF calf [3]. Subsequently, ultrasound-guided transvaginal oocyte aspiration was developed in the human [4,5] and then adopted in the bovine in 1987 in Denmark [6], to collect oocytes transcutaneously after hormonal pre-stimulation of heifers, and at the University of Utrecht, where Theo Kruip was part of the team [7], to collect them transvaginally from cows without hormonal stimulation. The current technology of OPU/IVP aims at harvesting (immature) oocytes from preselected genetically superior living cows by ultrasound guided transvaginal aspiration, followed by in vitro maturation, fertilization and culture until embryos have reached the morula or blastocyst stage and can be transferred

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