

1

Global benchmark in production

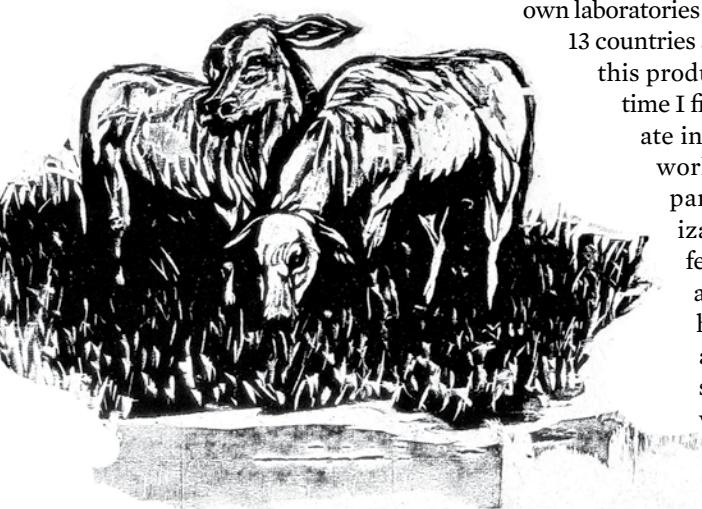
Investment in research and innovation by In Vitro Brasil accounted for 45% of the total world production of bovine embryos in 2013

In Vitro Brasil, located in Mogi Mirim, São Paulo State, is the leading Brazilian producer of bovine embryos in the laboratory—and the major contributor to Brazil's stature as the world's benchmark in this field. Of the 546,628 embryos produced in 2013 worldwide, according to data from the International Society of Embryo Technology (IETS), the Brazilian company accounted for 266,000 of them, or 45% of the total; 8 of its own laboratories plus 23 affiliates in

13 countries are responsible for this production. "Up to the time I finished my doctorate in 2007 and started working for the company, in vitro fertilization had been offered mainly to elite animals, donors of high commercial and genetic value," says Andrea Basso, who has a PhD in

animal breeding from the University of São Paulo and is currently head of research at the company, which last year earned R\$15 million. This was because it was an expensive procedure, and the technology was still under development in Brazil. "At first, for example, it was impossible to collect oocytes and transport them for more than eight hours before reaching the laboratory. Today we can transport them for up to 24 hours." It was also impossible to freeze in-vitro-produced embryos, and currently the company freezes about 40% of its production.

Since its founding in 2002, In Vitro has always emphasized investing in technology. Since 2007, the commercial focus of the company has changed and has been overtaken by the demand for farm animal reproduction, especially in the dairy herd. "We did our first commercial project of producing embryos on a large-scale, which lasted two years," says Basso. "Ultimately, we managed to produce 9,000 females of the Girolanda breed, the result of a crossing between the more productive Dutch breeds and the more hardy Gir



breed." Since completing this project in 2009, the company has carried out other commercial projects producing embryos for the dairy herd and beef cattle. "When farmers select in vitro fertilization, they can anticipate a genetic improvement of the herd for generations, both in dairy and beef cattle, which increases productivity."

At the same time the company also began working with scientific projects, including one using mass spectrometry to evaluate how long it was feasible to keep the embryo culture media under refrigeration. The project, which was supported by FAPESP through the Innovative Research in Small Businesses Program (PIPE), in collaboration with the Thomson Mass Spectrometry Laboratory of the University of Campinas (Unicamp), showed that they were more stable than imagined and enabled great advances in the in vitro production of bovine embryos. "Before the project, we made culture media once a week, but now we are producing them once a month and doing so in large quantities." She notes that when stored in a refrigerator, their shelf life extends to more than 60 days.

After this first project, another four were approved for the company under the same program. One of them, which ended last year, deals with molecular markers of fertility in the Girolanda breed. "We identified some regions of bovine chromosomes that could be candidate markers of fertility by measuring the conversion rate of oocytes in the embryos," says Basso.

Currently, the company has three other projects in progress. One project aims to establish a health certification protocol for Brazilian in-vitro-produced embryos, the ultimate goal of which is the export market. "If we can prove that embryos produced in the laboratory are free of pathogenic contaminants, we will have a tool to ensure quality and open the way to that market."

The second project involves developing a protocol to biopsy embryonic cells while preserving the viability of the embryo and, at the same time, provide sufficient material to do a genomic analysis of its characteristics. This project is a collaboration with São Paulo State University (Unesp), Araçatuba and Jaboticabal campuses, and with another livestock company Agropecuária Jacarezinho. "The company is working on a technology in which we are able to do a biopsy of the newly produced embryos in the laboratory, before they are frozen, and a genetic analysis so that the farmer can select which embryo is to be transferred to the recipient," says Basso. In a dairy herd, for example, the choice may be to improve the rate of fat or milk protein.

The third project intends to produce embryos of the Nelore and Dutch breeds from 2 to 5

1 Bovine embryos after six days of development

2 Manipulation of embryos in the laboratory



months old that have not yet entered puberty. The results have shown that prepubertal calves and heifers receiving certain hormonal stimulation were able to produce viable ova and embryos that could be transferred to recipients.

In Vitro still has two ongoing social projects, one funded by Sebrae, which supports micro and small businesses, in the northeastern state of Alagoas to improve the dairy herd. The company sent milk producers Girolanda embryos resulting in about one thousand positive pregnancies. Once the calves are born, the herd will see increased production, reaching perhaps 20 liters of milk per day; prior to the breeding project this number was between three and five liters. The other social project, which also aims to improve milk production, is being done in partnership with the company Zambezia Agropecuária of Mozambique, Africa. ■

Projects

1. Validation of biopsies and DNA amplification of in-vitro-produced bovine embryos for whole genome analysis (No. 2014/50616-0); Grant mechanism: Innovative Research in Small Businesses Program (PIPE); **Principal investigator:** Andrea Cristina Basso (In Vitro Brasil); **Investment:** R\$101,052.74 and \$4,745.00 (FAPESP).

2. Standardization and validation of PCR [polymerase chain reaction] in real time for health certification of in-vitro-produced bovine embryos (No. 2014/50169-4); Grant mechanism: Innovative Research in Small Businesses Program (PIPE); **Principal investigator:** Juliana Hayashi Tannura (In Vitro Brasil); **Investment:** R\$137,675.00 (FAPESP).

3. Development of genetic tools for selection of breed cows presenting high fertility (No. 2012/51067-5); Grant mechanism: PIPE; **Principal investigator:** Andrea Cristina Basso (In Vitro Brasil); **Investment:** R\$229,232.80 (FAPESP).