BOVINE EMBRYO PRODUCTION IN BRAZIL: A NEW SCENARIO

Viana, J.H.M.; Camargo, L.S.A.

Embrapa Gado de Leite, Rua Eugenio do Nascimento 610, Juiz de Fora, MG, 36038-330, Sociedade Brasileira de Tecnologia de Embriões, jhmviana@cnpgl.embrapa.br

Abstract

Bovine embryo transfer in Brazil begun in the 70´ies, first using embryos imported and latter collected in the country. The activity developed gradually, gaining an increasing economic importance. In the last six years, the incorporation of the laboratory embryo production technology, known as in vitro fertilization (IVF) or in vitro embryo production (IVEP) made a revolution in the ET market in Brazil. Embryo production increased significantly and the country becomes the largest bovine embryo producer in the world, reaching approximately to ¼ of the total of transfer performed. The activity focused in zebu and beef breeds, with embryos transferred mainly without cryopreservation. After a period of growth in both techniques, the IVF started to dominate the conventional ET market. Among factors which allowed this change are some particular characteristics of the Brazilian beef market and of the zebu breeds reproductive physiology, creating a contrasting situation with the world scenario. The use of reproductive biotechnologies is well established in Brazil, and will be of great importance for the national bovine germoplasm market. **Key-words**: Embryo transfer, in vitro fertilization, statistics, bovine, Brazil

Introduction

Bovine embryo transfer (ET) in Brazil begun in the 70´ies, when the first births of calves generated from frozen imported embryos and, soon latter, from embryos collected and transferred in the country were registered. The ET industry shows, therefore, almost 30 years in Brazil, as reported in the Celebrating Meeting of the 20 years of the Brazilian Embryo Transfer Society (Rubin, 2005). In this period, significant changes occurred in the activity, including technological, conceptual or of economic impact for cattle industry. New biotechnologies were developed and incorporated, extending the significance of ET (initially restricted to embryo production by conventional superovulation, or *in vivo*), and resulting in the change of the word “transfer” to “technology” in the SBTE name. The paradigm of an activity elitist and unplugged from cattle industry reality was changed to a highly demanded tool by the productive sector, and essential for bovine genetic improvement programs. From technology importer, Brazil becomes a world reference in this area. But there is no better example of this revolution than the increase of *in vitro* embryo production and the subsequent change in the market observed in the last few years.

By 2000 Brazil already has an expressive participation in bovine world embryo production. With 69.400 transfers communicated, the country was responsible for 13.13% of all transfers and to 82.11% of those performed in South America, being individually and outside the axis USA-Canada the country with the greater bovine embryo production (Thibier, 2001). In 2000, however, there was a particularly relevant fact: it was the first year in which Brazil communicated an expressive number (12.597) of transfers of embryos produced by *in vitro* fertilization (IVF). This year marks the beginning of the commercial success of this technique in the country, in which the use of IVF was until then restricted to academic area. *In vitro* fertilization was, at that time, an already known and applied to farm animals technique – the first birth of a calf generated by IVF occurred in 1981 (Brackett et al., 1982), almost 20 years before. But the association of the state of the art of *in vitro* embryo production (including all procedures, from maturation to the final steps of culture) with market conditions, restricted commercial application of IVF in the decades of 80 and 90. One of the breakpoints of this scenario was the development of ovum pick-up technique (OPU), necessary to make oocyte recover from live donors viable, and which only occurred in the last 80´ies (Pieterse et al., 1988). This technique, however, was initially focused in cows with acquired fertility problems (Looney et al., 1994), and only latter seen as an IVF routine tool (Galli et al., 2001).

The evolution of the IVF in the period 2000 - 2003

Since 2000 a continuous and expressive increase on *in vitro* embryo production was observed in Brazil (Table 1). This increase was, initially, associated with an increase in the whole embryo activity, once the conventional ET was also expanding. Among the facts which contributed to this scenario, one of the most important was the beef market situation. With more than 180 million animals and only 7% to 8% of the herd inseminated (Nehmi, 2004), there was a demand of bull for natural mating estimated in more than one million animals in Brazil. The possibility of multiply selected genotypes by embryo production benefited the zebu elite genetic market and, at the same time, made viable the dissemination of these animals in commercial herds, in which the necessity of productivity gains was pushed by the bad relationship between supplies needed and meat prices. The ET market, emerged in dairy and European breeds (Rubin, 2005), moved to beef and zebu breeds, with Nelore being responsible for more than 70% of the transfers in 2003 (ABCZ, 2004).

Table 1: Transfer of embryos produced by conventional superovulation (ET) and *in vitro* fertilization (IVF) in Brazil, in the period of 2000 to 2006

	Year							
Technique	2000	2001	2002	2003	2004	2005	2006	Variation
IVF	12,597	401	48,387	63,064	80,833	128,914	196,663	↑ 1,561%
ET	38,595	46,301	79,295	111,424	102,100	107,717	69,886	↑ 181%
Total	51,192	46,702	127,682	174,488	182,933	236,631	266,549	↑ 521%

The Brazilian situation in the world bovine embryo market

The increase of *in vitro* and *in vivo* embryo production in Brazil contrast with actual world scenario, in which countries or regions with expressive participation in the market in the past now show stagnation or even retraction in the activity. The number presented by the International Embryo Transfer Society (IETS) show that the unique region in which the ET industry had a similar trend to Brazil was Asia, mainly the Popular Republic of China (Thibier, 2006). In this country, however, the context is far different from Brazil, with the transfer of imported frozen embryos and, in the IVF, of oocytes recovered from slaughterhouse ovaries. In the Europe and USA the *in vitro* embryo production represented, in 2005, to only 2.18 and 0.5% of world total, respectively. Consequently Brazil was responsible, in the last four years, to approximately 25% of total embryo transfers and to half of IVF bovine embryos produced in the world (Table 2). The retraction in IVF industry in Europe and USA seems to be consequence of a number of reasons which include the low results of the technique in *Bos taurus*, sanitary problems such as foot and mouth disease and BSE, reduction in exports of bovine germoplasm and, mainly in Europe, in the lower interest in productivity gains in the herds.

Table 2: Participation of Brazil in the world embryo transfer activity

	In vivo (TE)		*In vitro* (IVF)		Total	
Year	Brazil	% World	Brazil	% World	Brazil	% World
2005	107,717	17.6%	128,914	48.46%	236,631	26.9%
2004	102,100	18.6%	80,833	33.7%	182,933	23.2%
2003	87,732	16.22%	63,164	59.5%	150,896	23.3%
2002	110,376	20.5%	48,670	58.4%	159,046	25.6%
2001	46,301	10.2%	401	1.3%	46,702	9.7%

The inversion in the proportion of *in vivo* and *in vitro* produced embryos after 2003

After 2003, a change in the national embryo market is observed (Figure 1), characterized by the retraction in embryo production by conventional superovulation and replacement by *in vitro* embryo production, which is responsible, since 2005, for the greater part of the transfers (Viana, 2006). Despite some economic reasons, such as the cattle industry crisis of 2004-2005, may have contributed to this change, the success of *in vitro* embryo production in Brazilian conditions is evident. Factors like the large size and high fertility of the herds, availability of recipients for fresh transfer, the market value of elite animals, the interest in calves of both gender, the presence of a well established market of medium and disposable material suppliers and the good receptivity of new technologies by the productive sector were determinant for commercial scale viability of IVF. The existence of expert technicians, as demonstrated by the growth of SBTE and its scientific production, was also important. In 2005, the in vitro embryo production (including OPU, maturation, fertilization and embryo culture) was the main theme of 31.54% of the abstracts presented (Viana, 2005).

One factor which must be highlighted, however, are the differences observed in many parameters of ovarian physiology in zebu cattle, including in the number of follicular waves, divergence, maximum size, persistency of dominant follicles, follicular recruitment, etc. (Rhodes et al., 1995; Figueiredo et al., 1997; Viana et al., 2000; Sartorelli et al., 2005). These characteristics seems to be associated with the great variation in superovulatory responses observed in zebu cattle (Baruselli et al., 2006), but also determinate the greater number of small growing follicles present in the ovaries and, consequently, a greater number of COCs recovered by OPU (Viana et al., 2004), when compared to results of *Bos taurus* (Wagtendonk-de Leeuw, 2005). Considering that fertilization, cleavage, blastocyst production and pregnancy rates are quite similar among laboratories all over the world; differences in the number of COCs recovered from each donor may be determinant to the viability or not of IVF. The follicular recruitment repeatability (Boni et al., 1997) and the less complex logistic in oocyte recover, when compared to ET, also contributed to make the IVF more competitive.

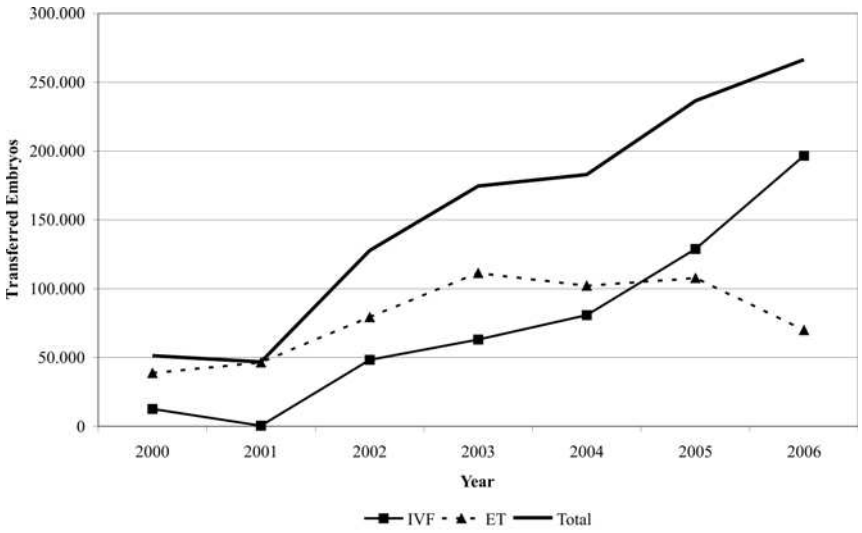

Figure 1: Transfer of embryo produced by conventional superovulation (ET) or *in vitro* fertilization (IVF) in the period of 2000 - 2006 in Brazil.

The scenario today

The trends observed in 2004 and 2005 were consolidated in 2006, with IVF reaching the expressive level of 73.8% of reported transfers in the country. Computing all transfers recorded (266.549), Brazil will probably keep its position as a world leader in bovine embryo production. The expansion of IVF established or increased some characteristics of the national ET market, such as the predominance of zebu beef breeds, associated with more than 90% of the transfers in 2006 (Table 3). The lower percentage of transfers in *Bos taurus* breeds also reflects the reduction in the use of imported embryos, which reached 15.3% of the transfers of frozen embryos and 3.6% of the total in 1996, and was reduced to an inexpressive value today.

Other characteristic of the market today is the preference for transfer of embryos without cryopreservation (Table 4), both in conventional ET (>80%) and in IVF (>95%), which reflects the low results obtained with frozen embryos from zebu breeds and produced *in vitro*. The preference for fresh transfer increased the recipient market, including the supply or housing of animals, estrous synchronization, pregnancy diagnosis and fetal gender identification.

The veterinary service in this area followed the changes resulting from the increase in IVF industry. A greater segmentation of the activity was observed, with procedures such as OPU and embryo transfer, formerly supplied by the IVF laboratories, being incorporated to the routine of professionals who worked before only with conventional ET. The centralization of actions typical of *in vivo* embryo production was replaced by the association of different expertise in the same process, with a higher degree of specialization in each part of the activity.

Table 3: Transfer of embryo produced by conventional superovulation (ET) or *in vitro* fertilization (IVF) in Brazil in 2006, stratified by breed group.

Breed group	ET	IVF	Total
Zebu dairy breeds	6,037	13,182	
European dairy breeds	2,615	0	
Subtotal	**8,652**	**13,182**	21,834 (8.19%)
Zebu beef breeds	59,568	183,481	
European beef breeds	1,666	0	
Subtotal	**61,234**	**183,481**	244,715 (91.81%)
Total	69,886 (26.22%)	196,663 (73.78%)	**266,549**

Table 4: Production, transfer and cryopreservation of bovine embryos in 2006.

	ET			IVF		
Breed	Collected	Transferred	Frozen	Produced	Transferred	Frozen
Dairy	12,052	8,652	2,600 (23.1%)	14,290	13,182	1,016 (7.2%)
Beef	69,062	61,234	6,954 (10.2%)	190,043	183,481	6,562 (3.5%)
Total	81,114	69,886	9,554 (12.0%)	204,333	196,663	7,578 (3.7%)

Embryo production in dairy breeds

Embryo production in dairy breeds by conventional superovulation fluctuated in the last 10 years, without expressive numeric changes (Table 5). Considering the whole activity, however, participation of dairy breeds was reduced, dropping from 36.9% in 2000 to less than 4% in 2004, and showed a trend of increasing since 2005. The participation of dairy breeds in the IVF market, few expressive up to 2004, showed a consistent increase after 2005 in zebu breeds, overtaking conventional ET in 2006 (Table 3). The use of IVF in dairy herds was limited by the low scale, consequence of herd sizes, by the lower fertility inherent of lactating cows, by the high male/female proportion in born calves, and by the low profitability of dairy industry. The increase observed in the last two years reflects a warm up in milk market, associated with a scenario of higher competition among beef breeds and possibly to the opportunity of using the already established capacity of the labs. The availability of the facilities and staff creates the opportunity to perform IVF in breeds or crossbreeds with lower market value and initially less attractive. In 2006, for example, the transfer of 2,580 ET embryos and 3,200 IVF embryos in Girolando breed (Gyr vs. Holstein) was reported. Another great impact factor was the introduction of sexed semen in the ET and, mainly, IVF routines (Dell´Aqua Jr. et al., 2006). The poor results obtained with frozen *in vitro* produced embryos, however, is still an important obstacle for IVF growth in dairy breeds, once the need for cryopreserved embryos in these breeds is higher (Table 4).

Table 5: Participation of dairy breeds in the total of conventional embryo transfers in the period 1997-2006

	Year									
	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006
Total	13,724	43,245	74,067	38,595	46,301	79,295	111,424	102,100	107,717	69,886
Beef	12,631	28,423	59,137	24,363	*	68,363	105,364	98,117	101,986	61,234
Dairy	1,093	14,822	14,930	14,232	*	10,932	6,060	3,983	5,731	8,652
(% total)	7.96%	34.27%	20.16%	36.88%		13.79%	5.44%	3.90%	5.32%	12.38%

* Lost data

Conclusions

Although it is not possible to predict the future evolution of ET scenario in Brazil, some trends seems well defined. The IVF is established as an option for multiplication of high value animals. Due to its high production capacity, after providing embryos for internal market the IVF will probably focus embryo export, mainly because of the country bovine genetic, adapted to tropical environment. This will require an extra effort in overcoming some technical limitations of IVF, including cryopreservation. The technology will also be applied to other farm species, as equine and small ruminants. Job opportunities may increase, including by the export of services and technology, but will require a higher professional qualification. Conventional TE, by the other hand, will remain important, but focused in more specific market opportunities. Finally, we must emphasize that IVF was an example of new biotechnique which was quickly incorporated to the production chain and resulted in significant advance for Brazilian cattle industry, demonstrating the importance of investments in research and technological development.

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