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MALFORMATIONS FOUND BY AUTOPSY OF CLONED AND TRANSGENIC PIGLETS OF DIFFERENT BREEDS

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Introduction and aim

Viability is seriously compromised in a high proportion of cloned and transgenic piglets, and one obvious reason is malformation of vital organs. In this study, malformations were described in dead pre-weaned piglets born after transfer to Large White (LW) recipients of cloned (LW donor cells) or transgenic (Yucatan or Göttingen donor cells) embryos.

Materials and methods

Donor cells were fibroblasts, with LW cells from LYxD, and with Göttingen and Yucatan cells made transgenic with one of five genes related to different human diseases. Handmade cloning was used to produce embryos that after 5 to 6 day's *in vitro* culture were transferred to 145 LW sows on day 4 after natural heat. Of these, 28 sows delivered cloned LW piglets, while 21 and 25 sows delivered transgenic Göttingen and Yucatan piglets, respectively. Malformed piglets were sacrificed, and the stillborn and dead pre-weaned piglets were autopsied to register possible malformations. Data were analyzed by Fisher's Exact test with a significance level of $P<0.05$.

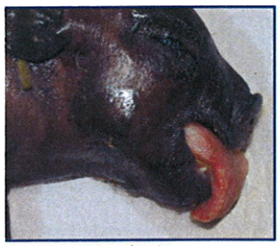

Figure 1. Macroglossia

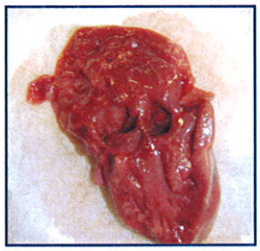

Figure 2. Heart septum defect

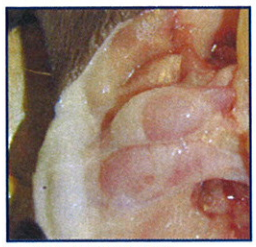

Figure 3. Cryptorchidism

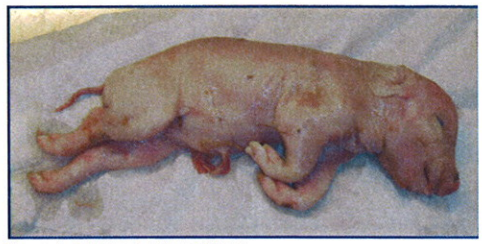

Figure 4. Joint contracture of legs

Results

In the 74 litters, total litter size ranged from 1 to 22 piglets (mean 6.3±0.5), and the overall mortality rate until weaning was 56%. Malformations were found in piglets from 55 litters (Table 1 and Figure 1-4). In those litters, 1-7 piglets had one (n=123), two (n=22) or several (n=16) malformations. The malformation rates of all born piglets in the transgenic Göttingen (38%) and Yucatan (33%) piglets were significantly higher than in the cloned LW piglets (21%). Some of the malformations seemed to be related to breed and/or transgene; for instance were heart malformations most frequent in Yucatan litters independent of the transgene, and gall bladder and gonad malformations were more frequent in various litters with the same transgene (Figure 5).

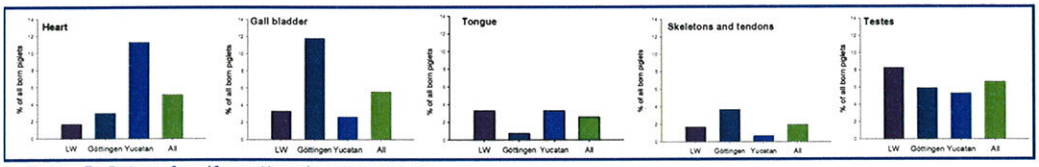

Figure 5: Rates of malformations in some organs

Discussion and conclusion

These results show that the use of cloning in pigs results in a considerable loss of piglets due to malformations. Use of transgenic cells for cloning only adds to this problem. This highlights the serious challenge it is to use pigs as model animals for human diseases, and the choice of breeds and transgene for this kind of work should be considered carefully. However, the large variation in the results illustrates that further improvements in production of cloned/ transgenic embryos are possible and may ultimately reduce the overall incidence of malformations.

No of autopsied /no of all born	No of litters with malformed /all litters	Number of piglets with one major malformation in these organsystems:									No of malformed piglets	% malformed piglets of all born
		Skeleton, muscles, diaphrag.	Kidneys and urinary tr.	Tongue	Heart	Testis	Brain and spinal cord	Liver and gall bladder	Intestines and pancreas	Several malforma tions		
LW n = 56/182	17/28	3	3	6	3	15	0	6	0	2	38	[a] 21%
Göttingen n = 67/136	20/21	9	1	1	4	8	0	16	3	10	52	[b] 38%
Yucatan n = 67/145	18/25	1	4	5	17	8	3	4	2	4	49	[b] 33%
All n = 190/463	55/74	13	8	12	24	31	4	26	5	16	139	30%

Table 1: Malformations in autopsied piglets born until 1.10.2011 (i.e. deviating from numbers in the abstract). $P<0.01$ in rows with different superscript.

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