Management of Newborn Triplets in a Marmoset Breeding Unit: **Example of a Successful Approach**



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Introduction

In captivity, marmosets very often give birth to triplets or more, unlike in the wild (on average 2 infants per litter). However, as females only have two teats, they can properly care for only two newborns. As a cooperative breeding species, the male and the juveniles from previous litters also contribute to raising the infants. However, this is not enough to compensate for the limited milk production. There are several protocols describing ways of managing surplus newborns beyond 2 per litter (natural selection, bottle-feeding, euthanasia, etc.). In this project, we carried out a pilot study to optimize the practice of bottle-feeding (nursing) by rotating babies, before deploying a new decision-tree more widely. This protocol is a good example of the implementation of a 3R approach in a non-human primate breeding center.

Material and methods

Animals

Housed in family groups until 8-12 months old, then in single-sex social groups. Standard housing conditions compliant with local regulation and AAALAC certified.

Little size can vary from 1 to 5 infants:

Litter size	ERBC Toulouse data	Reference ^a	
1	6.89 %	7.48 %	
2	26.48 %	37.41 %	
3	59.37 %	41.34 %	
4	6.53 %	4.28 %	
5	0.73 %		

^aMoyenne de: Tardif et al., 2003; Ash & Buchanan-Smith, 2014; Bakker et al., 2018: Box & Hubrecht, 1987; McCoy et al., 2019.

Decision-tree for managing newborns

- 1. Birth weight < 28g: euthanasia of the smallest (anesthesia overdose under deep sedation);
- 2. Birth weight > 28g and possibility for fostering: transfer according to
- 3. Birth weight > 28g without possibility for fostering: nursing protocol below.

Equipment and scheduling for nursing

- · Incubator with separators
- Heating mat
- Fleece and/or plush · Scale
- · Formula milk for babies
- Water bath
- Svringe
- · Gauze



Age (d)	Number of infants fed per day	Monday to Friday		Saturday/Sunday/ Public Holiday			
		Number of intake per day	Feeding hours	Quantity per intake (mL)	Number of intake per day	Feeding hours	Quantity per intake (mL)
1-7	1	4	09:00 / 11:30 14:00 / 16:30	0.5 – 1.5	3	08:30 / 10:30 12:30	0.5 – 1.5
8 – 14	1	4	09:00 / 12:00 15:00 / 17:00	1.5 – 3.0	3	08:30 / 10:30 12:30	1.5 – 3.0
15 – 21	1	3	09:00 / 12:00 16:00	2.0 – 5.0	2	08:30 / 12:30	3.0 – 5.0
22 – 30	3	2	10:00 / 16:00	3.0 – 5.0	1	11:00	3.0 – 5.0

Nursing protocol

- Prepare formula milk as supplier instructions, keep at good temperature in
- Take the litter our of the family and weigh them. Keep one for nursing.
- 3. Check the milk temperature on one's wrist.
- 4. Slowly feed with the syringe, at Nourrir lentement avec une seringue, following the swallowing pace.
- 5. Once full, stimulate the anogenital area with a warm gauze to encourage urination and defecation.
- 6. Place the baby in the incubator set to 30°C and repeat the operation at each hour indicated in the schedule.
- 7. At the end of the day, return the infant to its family.
- 8. The following day, proceed in the same way with another baby. On day 3, take the 3rd one and continue this way until day 30.

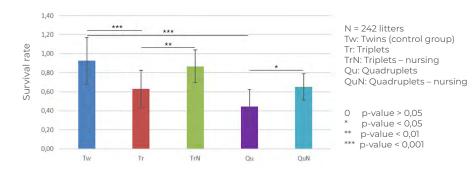
This protocol was initiated by the facility Animal Welfare Body and supported by the Ethics Committee in Animal Experiments.

2 Results

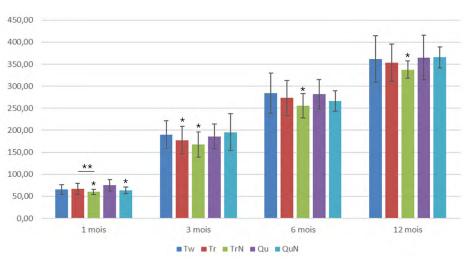
Survival rate at 1 month

Without nursing: euthanasia of the surplus newborn(s)

- + 37.3 % for triplets
- + 46,9 % for quadruplets



Weight difference at 1, 3, 6 and 12 months				
1 month	Triplets and Quadruplets smaller than Twins			
	Triplets with nursing smaller than Triplets without nursing			
3 month	Triplets smaller than Twins and Quadruplets			
	Nursing and without nursing are equivalent			
6 and 12 months	Triplets with nursing smaller than Twins and Triplets without nursing			



N = 242 litters Tw: Twins (control group) TrN: Triplets - nursing Qu: Quadruplets QuN: Quadruplets - nursing 0 p-value > 0.05 p-value < 0,05 *** p-value < 0,001

This nursing protocol significantly increases the survival rate at 1 month for triplets and quadruplets.

The weight at 1 month is lower for infants that completed the nursing protocol. A significant weight difference remains between twins and triplets that completed the nursing protocol.

No anomalies (abnormal or stress-related behavior) were observed by the staff. To be confirmed with more exhaustive behavioral studies.

Conclusion

- · This protocol is very time-consuming but highly rewarding for the staff.
- Ethical value and refinement: less animals are euthanized and need for less breeders to obtain the same number of animals.
- · This protocol can be completed with behavioral and physiological data to evaluate its impact on other aspects.

Aknowledgement

We would like to thank all the ERBC Toulouse staff involved in this protocol, who contribute every day to keeping the animals in the best possible animal welfare conditions.

