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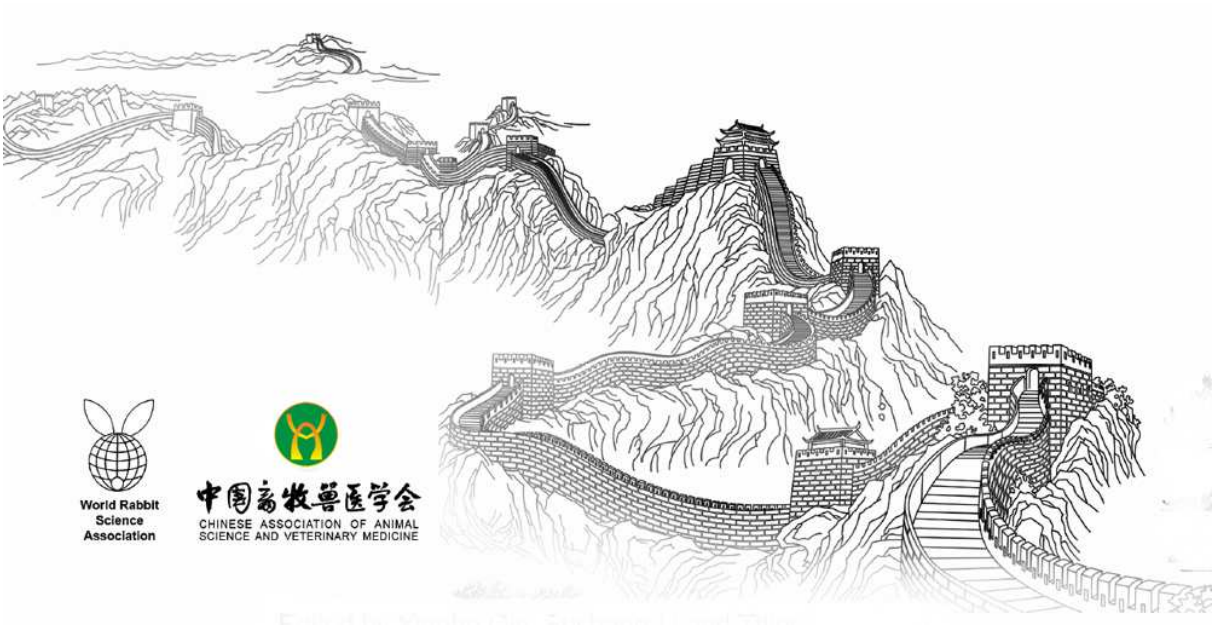

EFFECT OF A PLANT EXTRACT ON RABBIT EMBRYONIC VIABILITY

Bebin K.[1], Destombes N.[2], Robert R.[3] , Fournier E.[3], Briens C.[1]., Gardan-Salmon D.[1]

[1] CCPA Group - ZA du Bois de Teillay - 35150 Janzé, France.
[2] TERRENA – La Noëlle - 44150 Ancenis, France.
[3] EUROLAP – Le Germillan - 35140 Gosné, France
Corresponding author: kbebin@groupe-ccpa.com

ABSTRACT

Prolificacy is one of the key factors in successful rabbit farming. A better knowledge of state and nature of mortality during pregnancy according to genotypes is of great interest for breeders in order to improve genetic breeds. The aim of this study is to evaluate the effect of *Scutellaria* plant extract (SC) on embryonic viability in order to improve doe's prolificacy. Forty lactating multiparous Hyla rabbits were used in this study. Twenty does received Scutellaria plant extract supplementation (SC group), 20 others received the same feed without SC (control group). On day 15 of gestation, the 40 does were autopsied for diagnostic of gestation in order to count and assess the viability of the foetuses. Number of corpora lutea and implanted vesicles was not significantly different between the two groups. After opening vesicles, there were 1.6 times more resorbed or dead vesicles in control group than in SC group. Late mortality, i.e. between implantation and day 15 of gestation, was calculated at 7.5% in control group vs 5% in SC group. Necropsies of pregnant rabbit does are of great interest in order to get a better knowledge of stage and nature of mortality during pregnancy. Scutellaria administration in early gestation may have beneficial effects on initiation of placentation and vascularisation, leading to a higher embryonic survival.

Key words: Rabbit, embryonic viability, gestation diagnostic

INTRODUCTION

Prolificacy is one of the key factors in successful rabbit farming. In the field of research, it is therefore necessary to know and understand the components involved in this prolificacy, from ovulation to kindling. The chronological sequence of the various stages, from ovulation to kindling, is presented in Figure 1 (Laborda-Vidal 2011, Llobat-Bordes 2012, Caron *et al.*, 2012).

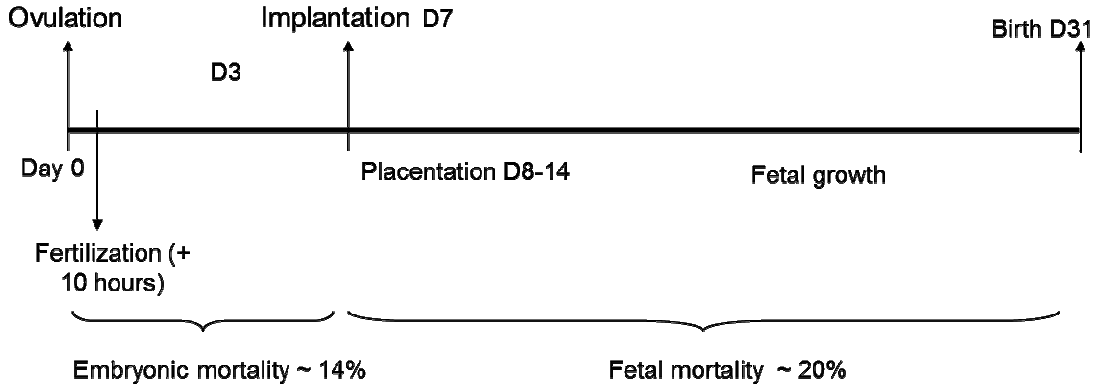

Figure 1. Embryo mortality during pregnancy in rabbits (Laborda-Vidal, 2011 and Llobat-Bordes, 2012).

The early events of pregnancy are associated with rapid changes in expression of genes for nutrient transport, cellular remodeling, angiogenesis, and relaxation of vascular tissues, as well as cell proliferation and migration (Bazer et al., 2009). Failure of conceptuses to undergo implantation and/or cell death during the peri-implantation period results in early embryonic loss. Those early embryonic losses have been evaluated to be responsible for 14% of mortality during pregnancy in rabbit (Laborda-Vidal, 2011) although some

differences can be observed between genetics. A better knowledge of state and nature of mortality during pregnancy according to genotypes is of great interest for breeders in order to improve genetic breeds. Necropsies of gestating rabbit does allow discrimination between early embryo losses (i.e. between ovulation and implantation) or late embryo losses (after implantation), which is not possible to evaluate with other non-invasive techniques.

Enhancement of placental growth and function through nutritional management offers an effective solution to improving embryonic and fetal survival and growth. Scutellaria plant extract has been shown to have protective effects against implantation failure in mice by anti-inflammatory effects or by reducing high oxidative stress (Ma et al., 2009; Zhao et al., 2011). Scutellaria plant extract also have angiogenesis properties (Zhang et al., 2011). We hypothesized that Scutellaria administration in early gestation could have beneficial effects on implantation and initiation of placentation, leading to a higher embryonic survival. Therefore, the aim of this study is to evaluate the effect of Scutellaria plant extract on embryonic viability in order to improve doe's prolificacy.

MATERIALS AND METHODS

Animals and insemination methods

Animals were reared in a conventional French farm. Forty lactating multiparous Hyla rabbits were inseminated 11 days after kindling. Fifty-four hours before insemination, a subcutaneous injection of 25 IU of eCG was administered. At the time of insemination, an injection of GnRH analogue (0.2 mL buserelin, Receptal, INTERVET, Angers, France) was administered to trigger ovulation.

Feeding method and light

The females were fed *ad libitum* with the same feed (2 500 kcal of digestible energy, 17.5 % of crude protein).

Three days after insemination, 20 does, randomised on the weight at insemination, received Scutellaria (SC) solid extract in a complementary diet during 5 days (SC group), 20 others received the same feed without SC (control group). The experimental diet had the same nutritive values and raw materials that the basal diet. The does received 50 g per day of this complementary feed.

Sacrifice and necropsy of animals

Sacrifices were conducted by electronarcosis under veterinary supervision. On day 15 of gestation, the 40 does were transported to the DELTAVIT laboratory for necropsy in order to count and assess the viability of the foetuses. Different measurements were taken: number of corpora lutea enabling determination of the number of eggs released, number of embryonic vesicles corresponding to the number of implanted embryos and number of viable foetuses (organogenesis accomplished and size in consistent with developmental stage) and non-viable foetuses (foetuses with incomplete development and size or haemorrhagic vesicle) (Figure 2). Early embryonic mortality was evaluated as the difference between corpora lutea (oocyte ovulated) and number of vesicles (embryos implanted). Late mortality was evaluated as the difference between number of vesicles and viable foetuses.

This study completes Robert et al. study on the diagnostic of gestation (Robert et al., 2015).

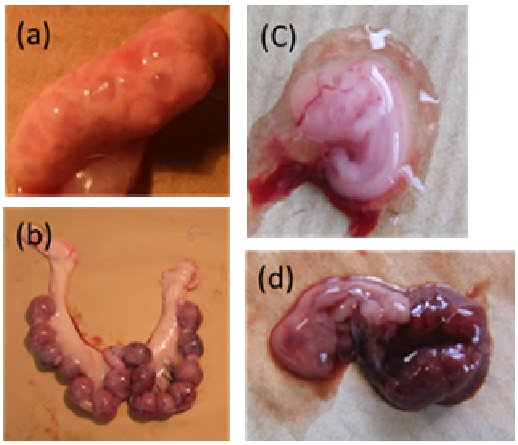

Figure 2. Illustrations of the observations performed during necropsy: (a) ovary for counting of corpora lutea, (b) isolated gestating uterus to identify and count embryonic vesicles, (c) viable foetus on day 15 of gestation and (d) haemorrhagic vesicle and non-viable foetus on day 15 of gestation.

Statistical analysis

Statistical analysis of data was conducted using SPSS 19.0; data were analysed with one-way analysis of variance (ANOVA). In all statistical tests, P values less than 0.05 were considered as significant

RESULTS AND DISCUSSION

Necropsies of pregnant rabbit does offer important information on ovulation capacity of the breeds and precise evaluation of embryonic mortality stages. In this study, similar number of corpora lutea was counted in control and SC groups, respectively (Table 1; P>0.1). Number of embryonic vesicles was not significantly different between the 2 groups. Early embryonic mortality (between ovulation and implantation) was evaluated at 11.5% in control versus 6.9% in SC group (Figure 3, P>0.1), which is in accordance with previously reported in rabbit (Llobat-Bordes, 2012). This data did not reach significance because of low number of does necropsied, but is a huge physiological difference between the 2 groups. After opening vesicles, there was 0.55 more resorbed or dead foetus in control than in SC group (Table 1). Late mortality, i.e. between implantation and day 15 of gestation, was calculated at 8.4% in control group vs 5.4% in SC (P>0.1).

Although number of vesicles implanted and number of live foetus was not different between the 2 groups (Table 1), viability of embryos seems to be improved with Scutellaria supplementation since less resorbed or dead foetus were observed. The total viability of foetus, i.e between insemination and day 15 of gestation, tended to be lower in the SC (11.94%) than in the Control group (19.25%) (P=0.1; Figure 3). Our hypothesis is that Scutellaria, through its angiogenic properties, could improve vascularisation of the implanted vesicle leading to better embryo survival.

Table 1. Effect of Scutellaria supplementation between insemination and implantation times on corpora lutea, embryonic vesicles and foetuses after necropsy on day 15 of pregnancy.

	Control group	SC group	SEM	P value
Corpora lutea	16.8	15.7	0.74	0.275
Embryonic vesicle	14.35	14.45	0.55	0.894
Fetuses resorbed	1	0.65	0.25	0.313
Dead fetuses	0.2	0.1	0.10	0.466
Live fetuses	13.25	13.8	0.62	0.509

Data are expressed as means (n=20). SEM: standard error of the mean.

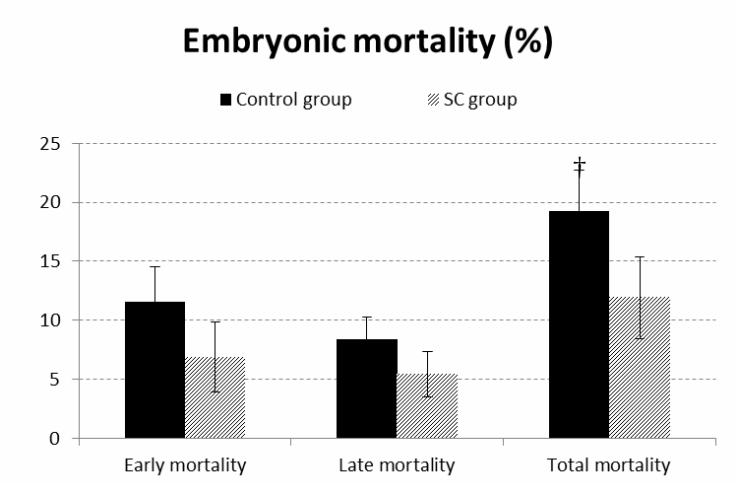

Figure 3. Effect of Scutellaria supplementation between insemination and implantation times on embryonic mortality (†, P=0.1). (n=20 does per group)

CONCLUSION

Our study provides new insights in embryonic viability evaluation in a commercial genotype of rabbit does. In this study, early embryonic mortality (before implantation) accounts for 9.2% of losses and late mortality (between implantation and day 15 of gestation) for 6.9%; which results in a global foetal mortality (from insemination) at 15 day of gestation of 15.6%. Those results confirm previous data from Fortun et al. (1993) and Laborda et al. (2012), even if prolificacy has increased. Unfortunately, our invasive method does not allow estimating late fetal mortality (from 15 days of gestation to birth). Although further studies are needed, supplementation with Scutellaria after insemination may be a promising strategy in order to help improving embryo viability and therefore prolificacy.

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