

Reproduction of the European hare in Patagonia, Argentina

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Monthly samples totalling 202 European hares *Lepus europaeus* Pallas, 1778 were collected in southern Argentina from August 1993 to March 1994. The sex ratio of the whole sample was 1:1.2 in favour of females. Pregnant females were present from August to February and lactating females were present from September to March. The percentage of pregnant females was 59% for the whole breeding season. The mean ovulation rate for female hares was 2.78; the number of ova ovulated was not significantly correlated with body weight. The mean number of implantation sites per reproducing female was 2.46. The number of embryos in a litter varied from 1 to 4, the average being 2.12. The prenatal mortality was 56.5% of all ova ovulated and of these 34.7% were lost before implantation and 21.8% after implantation.

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Introduction

In South America, the European hare *Lepus europaeus* Pallas, 1778 was first introduced in central Argentina and southern Chile. Its present range includes Uruguay, Paraguay, and southern parts of Bolivia and Brasil (Grigera and Rapoport 1983, Flux *et al.* 1990). In Argentina, the first hares were imported from Germany in 1888 and liberated in the Santa Fe province. At present, they occupies most grassland and agricultural areas over almost all of the country (Grigera and Rapoport 1983). The hares are regarded as a pest in Argentina and are favoured for sport and commercial hunting. In many areas the damage hares do to forestry plantations, crops, orchards, and in competing with domestic stock for grazing are considerable (Jackson 1988, Bonino 1995). On the other hand, it is probably the most important game animal in this country, where more than 6 million hares are shot annually for their meat which is exported to Europe (Fujita and Calvo 1981, Jackson 1986). The density of hares in different habitats of Patagonia range from 2/ha in the tussock grass steppe to 7–12/ha in the meadow valley (Novaro *et al.* 1992, J. Amaya *et al.*, unpubl.). In any case, the strategies for monitoring reproductive success and for the managing hare population will depend on knowledge of the reproductive characteristics of this species.

This paper describes a small study on sex ratio, female fertility, litter size, and prenatal mortality of the European hare in Patagonia, Argentina.

Study area

This study was conducted near Bariloche (41°10'S, 71°05'W), Río Negro province, Argentina. The area consists of a broad valley surrounded by mountains and with an ephemeral stream course. It is mainly sheep-grazing country where elevation varies from 800 to 1100 m above sea-level. The precipitation mostly occurs in the fall-winter season and the annual average for the period 1936–1988 was 690 mm. Annual average temperature is 7.1°C, with mean monthly extremes of 1.3°C in July and 12.8°C in January (Bustos and Rocchi 1993).

The study area lies within the Patagonic Phytogeographical Province described by Cabrera (1971). The vegetation consists mainly of grasslands dominated by *Stipa speciosa* var. *major* at the lower elevations and by *Festuca pallescens* at the highest elevations. An important shrub associated with these grasses is *Mulinum spinosum*. The valley bottom is vegetated by moist meadows of *Juncus balticus*, *Poa pratensis*, *Hordeum comosum*, and *Taraxacum officinalis*. Along the water courses there are shrubs such as *Berberis heterophylla*, *Discaria* spp., *Schinus patagonicus*, and *Maytenus boaria*. There are also remnants of mountain forest dominated by *Austrocedrus chilensis* and *Nothofagus antarctica*.

Materials and methods

Monthly samples totalling 202 hares were collected by shooting in the breeding season from August 1993 to March 1994. Each hare was dissected within 12 hr of death. Both eyes were removed and preserved in 10% formalin for about 7 days. The abdomen was opened after estimating the activity of the mammary gland. The testis of males and the ovaries and reproductive tract of females were excised and preserved intact in 10% formalin. In the laboratory the preserved uterus was opened and the number of embryos and implantation sites noted. The ovaries were cut freehand with a scalpel into longitudinal sections approximately 1 mm thick and examined macroscopically to count the corpora lutea. The diameter of corpora lutea was measured to the nearest 0.1 mm. The number of corpora lutea was used as an indication of the number of ova ovulated (Cheatum 1949, Kirkpatrick 1987).

Discrimination of age classes was carried out by means of dry eye lens weight (Caboń-Raczyńska and Raczyński 1972, Brandani *et al.* 1977, Suchentrunk *et al.* 1991). The animals were separated into 3 age classes: leverets (234.9 mg or lighter), juveniles (235.0–279.9 mg) and adults (280.0 mg or heavier). Juvenile and adult individuals of both sexes were considered reproducing in accordance with Amaya *et al.* (1979).

Results and discussion

Sex ratio

The material examined was composed of 92 males (21 leverets, 38 juveniles, and 33 adults) and 110 females (30 leverets, 40 juveniles, and 40 adults). Sex ratio for European hares collected during this study was 1:1.2 in favour of females. For the same region, Amaya *et al.* (1979) reported similar sex ratio (582 males : 711

females), whilst Bustos (1986) found that the ratio was 1:1.1 in favour of males (125 females : 139 males).

On nearly 180,000 hares examined in different countries of Europe, in all cases a slight predominance of females was found (Hell 1960, Jezierski 1965, Pielowski 1969) or the ratio was 1:1 (Raczyński 1964). In New Zealand, Flux (1967) found that in 1432 hares the ratio was 1:1.13 in favour of females.

Apparently the sex ratio is subject to certain fluctuations in different years, sometimes in favour of females, and sometimes in favour of males. This would be principally connected to method of capture and the reaction of individuals of the two sexes to shooting at different periods (Raczyński 1964, Pielowski 1969). For practical purposes this slight predominance of females is not important and a balanced sex ratio can be accepted in calculating the annual increase in the head of hares (Pielowski 1969).

Breeding season

The limits of the breeding season can be estimated from the monthly percentages of reproducing females pregnant (Fig. 1). Pregnant females were first found in August (all with resorbed entire litters) and last in February. On the other hand, the lactation period started in September and concluded in March. Although no specimens were collected it is not very probable that some females were capable of breeding before August since in this month no lactating females were detected. The end of female reproductive activity in February was assumed from the fact that we encountered no females that conceived after February. In all, the average of pregnant females was 59%. The incidence of pregnancy rose rapidly from 22% in August to 83% in December (Fig. 1). It declined after December to a low level (29%) in February. Despite the small samples, the level of reproductive activity in adult females was slightly higher than in juveniles (57% and

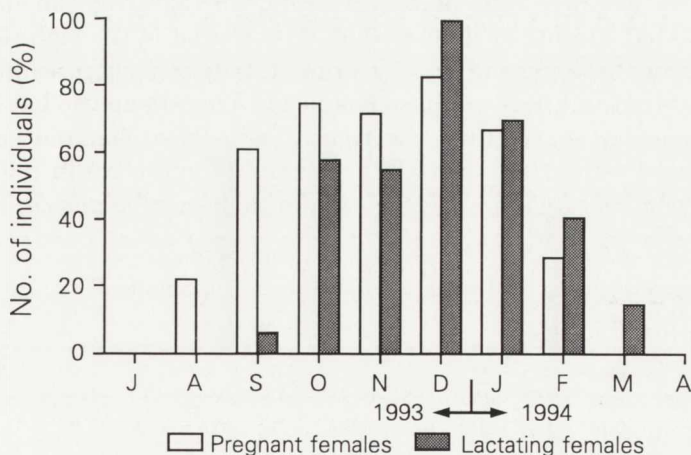


Fig. 1. Monthly percentages of pregnant and lactating females.

55%, respectively). Amaya *et al.* (1979) found a similar pattern in the breeding season for European hares collected on the same study area. *Lepus europaeus* starts breeding in all countries studied so far at approximately the same time of year regardless of environmental conditions (Flux 1965).

Female fertility

The number of ova ovulated indicating the potential litter size was defined indirectly by means of counting the number of corpora lutea in the ovaries. The total number of corpora lutea counted was 192. The number in a set varied from 1 to 8, 3 being the most frequent (Table 1). In all, the mean number of corpora lutea in a set was 2.78 with SD = 1.19; 3.13 ± 1.21 in the juvenile females ($n = 40$), and 2.46 ± 1.14 in the adult ones ($n = 40$).

The eight corpora lutea recorded for one hare may have included corpora lutea atretica or accessory corpora lutea which could not be distinguished because of the wide variation in size of the functional corpora lutea present. Flux (1967) encountered the same difficulty and different criteria of corpora lutea may well account for the wide variation in estimates of prenatal mortality reported by some authors.

During the breeding season the distribution of ovulation between the two ovaries was uneven, a greater number being ovulated from the left ovary in both age classes (juvenile and adult females). In 46 of the 69 does shedding more than one ovum both ovaries participate; in 14, ovulation occurred in the left ovary and in 9 in the right ovary only.

The number of ova ovulated was not significantly correlated with body weight of females ($r = 0.21$, $p > 0.05$) although this correlation is found in many species of mammals (Brambell 1944, Watson 1957).

Litter size

Based on 85 embryos and placental scars, 45 (53%) of the implantations occurred in the left uterine horn and 40 (47%) occurred in the right uterine horn. The difference in these percentages was not statistically significant. Valentincic (1956) reported that in European hare females in Yugoslavia the left uterine horn was more frequently gravid than the right, but in New Zealand embryos were evenly distributed with 210 in the right horn and 218 in the left in 180 pregnancies (Flux 1967). Tiemeier (1965) found the left uterine horn of *Lepus californicus* (602

Table 1. Frequency distribution and mean of corpora lutea, implantation sites and embryos in 80 reproducing female hares.

	0	1	2	3	4	5	6	7	8	Total	Mean
Corpora lutea	19	10	16	21	16	3	0	0	1	192	2.78
Implantation sites	37	15	23	10	5					111	2.46
Embryos	34	10	19	7	4					85	2.12

Table 2. Seasonal frequency distribution of litter size (number of embryos) in juvenile and adult female hares.

Month	Juvenile females						Adult females					
	0	1	2	3	4	Mean	0	1	2	3	4	Mean
August	7						6					
September	3	1	3			1.7	4	1	3			1.7
October	2	1	2	2		2.2	1	1	1		1	2.3
November	4	1	1			1.5		1	2	1		2.0
December	1	1		1		2.0		1	1	1	1	2.5
January		1	2	1	1	2.4	4	1	1	1	1	2.5
February			1			2.0	2		2			2.0
Totals	17	5	9	4	1	2.05	17	5	10	3	3	2.19

embryos) more frequently gravid than the right (535 embryos), and, according to Asdell (1946), it seems that is characteristic in several species of mammals. The number of implantation sites in a female varied from 1 to 4, 2 being the most frequent and 2.46 the average (Table 1). The number of embryos in a litter varied from 1 to 4, 2 being the most frequent (Tables 1 and 2). The mean litter size for all fertile females was 2.12. We found no significant difference between the mean litter size of juvenile females (2.05) and of adult females (2.19). The litters of juvenile were similar or slightly smaller than those of adults in all months (Table 2). Amaya *et al.* (1979, 1983) reported that mean litter size was 2.19 for European hares collected on the same study area, and 2.61 for hares collected in Santa Cruz province. Litter size in Bariloche (2.12) was not very different to the following litter sizes reported for other countries: 2.76 in Poland (Raczyński 1964), 2.21 in New Zealand (Flux 1967), 2.69 in Scotland (Hewson and Taylor 1975), 2.56 in France (Pépin 1989), and 2.73 in Slovakia (Ciberej 1993). Problems of reproductive physiology (superfoetation, polyovular follicles, migration of blastocysts, and ectopic pregnancy) reported by different authors (Raczyński 1964, Bloch *et al.* 1967, Flux 1967, Lloyd 1968) were not observed in this study.

Prenatal mortality

The prenatal mortality was estimated analyzing the data as described by Flux (1967). However, the numbers involved here are small and do not permit of critical analysis as do the data of Flux, therefore the results must be considered indicative of this mortality. Gross estimate of prenatal mortality are sometimes obtained by a simple comparison of the numbers of corpora lutea and healthy embryos. For 69 ovulating females a comparison of corpora lutea (192) and embryos (85) reveals a loss of about 55.7% of the ova. However, to obtain an adequate estimate of prenatal mortality it is necessary to consider pre- and post-implantation loss of entire litters, and of ova in litters which survive. Pre-implantation mortality

involves a loss of ova between the time of ovulation and the time of implantation. No attempt was made to recover ova from the uterine horns, and pre-implantation loss of entire litters could be estimate only by carefully examination of those females which had large corpora lutea but no implanted embryos. We assume that even at the newly implanted stage corpora lutea may reach 8 mm in diameter, as Flux (1967) states. Six of 80 (7.5%) fertile females in the pre-implantation stage had large corpora lutea (7–9 mm diameter) which seemed to be regressing, and these were considered females which had lost their entire litter before implantation. This represents an total pre-implantation loss of 3.7% since pseudo-pregnancy lasts about twice as long as the pre-implantation period (Lechleitner 1959). In this study, a total of 192 ova shed produced 111 implantation sites, indicating a loss of 81 ova (42.2%) in litters in which some ova survived to implant. This is considerably higher than the loss of 28.6% quoted for this species in New Zealand (Flux 1967).

A crude estimate of the losses after implantation can be made by comparing the number of implantation sites with the number of healthy embryos. In this study, 45 pregnancies show 6 litters (13.3%) with post-implantation loss and 26 of 111 (23.4%) of implantation sites show losses. One might assume that this was the total loss occurring after implantation but such an assumption is far from correct (Brambell 1942). It is necessary to estimate post-implantation loss of entire litters, and of ova in litters which survive. Post-implantation loss of entire litters was estimated comparing the number of litters in the sample with embryos younger (21) and older (18) than half term. The difference between these values gives a loss of three of 21 litters (14.3%) after implantation. The post-implantation loss of ova in litters that survive to full term may be calculated by comparing the number of implantation sites (23) and embryos (21) in 11 litters with embryos of 100 g or over. This represents a post-implantation loss of 8.7%. The whole of the loss can be calculated by converting all of the figures given previously into terms of ova shed. The loss of entire litters before implantation accounted for 3.7%, and 14.3% of the remaining 96.25% (13.8%) were lost entirely after implantation, giving a total loss of entire litters of 17.5%. The surviving 82.5% of the litters were subject to a 42.2% pre-implantation loss and 8.7% post-implantation loss. In terms of ova shed this was $[42.2 + (0.087 \times 100 - 42.2)] \times 82.5 = 39\%$, distributed between pre-

Table 3. Comparison of the percentage prenatal mortality in European hare as determined in this study with that determined by Flux (1967) in New Zealand.

Pre-implantation loss		Post-implantation loss		Total loss	Source
Entire litter	Ova in surviving litters	Entire litter	Ova in surviving litters		
2.3	21.9	14.3	9.0	47.5	Flux 1967
3.7	31.0	13.8	8.0	56.5	This study

and post-implantation in the ratio 42.2:8.7, ie 31% and 8%. Table 3 compares the losses estimated in this study with those determined by Flux (1967) in New Zealand, which shows that the losses were similar at every level, except that the pre-implantation loss of ova was higher in our case. Hewson (1964) estimated the partial mortality of litters in *L. europaeus* in Scotland by the difference in average size of litters with small embryos (3.19) and those with large embryos (2.46) to be 23%. Similar calculations with our data give a mean litter size of 2.09 for litters with small embryos and 1.65 for litters with large embryos, a partial mortality of 21%. On the other hand, Flux (1967) estimated a partial mortality of only 6% in New Zealand.

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References

- Amaya J., Alsina M. G. and Brandani A. 1979. Ecología de la liebre europea. II: Reproducción y peso corporal de una población de S.C. de Bariloche. Instituto Nacional de Tecnología Agropecuaria, Bariloche, Río Negro. Comunicación Técnica 9: 1–36.
- Amaya J., Bonino N., Clarke R. and Díaz M. 1983. Informe preliminar sobre la situación actual de la liebre europea *Lepus europaeus* en la provincia de Santa Cruz. Instituto Nacional de Tecnología Agropecuaria, Bariloche, Río Negro. Informe Técnico: 1–30.
- Asdell S. A. 1946. Patterns of mammalian reproduction. Comstock, New York: 1–437.
- Bloch S., Hediger H., Lloyd H. G., Muller C. and Strauss F. 1967. Beobachtungen zur Superfoetation beim Feldhasen (*Lepus europaeus*). Zeitschrift für Jagdwissenschaft 13: 49–52.
- Bonino N. 1995. Introduced mammals in Patagonia, southern Argentina: consequences, problems, and management considerations. [In: Integrating people and wildlife for a sustainable future. J. A. Bissonette and P. R. Krausman, eds]. Proceedings of the first International Wildlife Management Congress. The Wildlife Society, Bethesda: 406–409.
- Brambell F. W. R. 1942. Intra-uterine mortality of the wild rabbit *Oryctolagus cuniculus* (L.). Proceedings Royal Society, Serie B 130: 462–479.
- Brambell F. W. R. 1944. The reproduction of the wild rabbit *Oryctolagus cuniculus* (L.). Proceedings of Zoological Society of London 114: 1–45.
- Brandani A., Alsina M. G. and Amaya J. 1977. Ecología de la liebre europea. I. Estimadores de edad y estructura de una población de S.C. de Bariloche. Physis 36: 305–320.
- Bustos J. C. 1986. Variaciones en el peso y dimensiones del cuerpo y de ciertos órganos internos de la liebre europea (*Lepus europaeus*). Tesis de Licenciatura, Centro Regional Universitario Bariloche, Río Negro: 1–34.
- Bustos J. C. and Rocchi V. C. 1993. Caracterización termoplumiométrica de veinte estaciones meteorológicas de Río Negro y Neuquén. Instituto Nacional de Tecnología Agropecuaria, Bariloche, Río Negro. Comunicación Técnica 1: 1–43.
- Caboń-Raczyńska K. and Raczyński J. 1972. Methods for determination of age in the European hare. Acta Theriologica 19: 69–80.
- Cabrera A. L. 1971. Fitogeografía de la República Argentina. Boletín de la Sociedad Argentina de Botánica 14: 1–42.
- Casida L. E. 1960. Techniques and procedures in animal production research. American Society of Animal Science, New York: 1–228.
- Cheatum E. L. 1949. The use of corpora lutea for determining ovulation incidence and variations in the fertility of white-tailed deer. Cornell Veterinarian 39: 282–291.

- Ciberej J. 1993. Growth of embryos and losses during the prenatal development in European hare. *Folia Venatoria* 23: 99–112.
- Flux J. E. C. 1965. Timing of the breeding season in the hare *Lepus europaeus* Pallas, and rabbit *Oryctolagus cuniculus* (L.). *Mammalia* 29: 557–562.
- Flux J. E. C. 1967. Reproduction and body weights of the hare *Lepus europaeus* Pallas, in New Zealand. *New Zealand Journal of Science B* 10: 357–401.
- Flux J. E. C., Duthie A. G., Robinson T. J. and J. A. Chapman 1990. Exotic populations. [In: Rabbits, hares and pikas: status survey and conservation action plan. J. A. Chapman and J. E. Flux, eds]. Information Press, Oxford: 147–153.
- Fujita H. O. and Calvo J. O. 1981. Las exportaciones de productos y subproductos de la fauna silvestre en el quinquenio 1976–1980. *Idia* 397: 1–26.
- Grigera D. and Rapoport E. 1983. Status and distribution of the European hare in South America. *Journal of Mammalogy* 64: 163–166.
- Hell P. 1960. [Das Geschlechtsverhältnis beim Feldhasen (*Lepus europaeus* Pall.)]. *Zoologické Listy* 9: 83–88. [In Czech with German summary]
- Hewson R. 1964. Reproduction in the Brown hare and the Mountain hare in Northeast Scotland. *Scottish Naturalist* 71: 81–89.
- Hewson R. and Taylor M. 1975. Embryo counts and length of the breeding season in European hares in Scotland from 1960–1972. *Acta Theriologica* 20: 247–254.
- Jackson J. 1986. The hare trade in Argentina. *Traffic Bulletin* 7: 72.
- Jackson J. 1988. Terrestrial mammalian pests in Argentina: an overview. *Proceedings of Vertebrate Pest Conference, USA* 13: 196–198.
- Jezierski W. 1965. Studies on the European hare. VII. Changes in some elements of the structure and size of populations. *Acta Theriologica* 10: 11–25.
- Kirkpatrick R. L. 1987. Indices fisiológicos en la gestión de la vida silvestre. [In: Manual de técnicas de gestión de vida silvestre. R. Rodríguez Tarrés, ed]. Wildlife Society, Inc., USA: 105–118.
- Lechleitner R. R. 1959. Sex-ratio, age classes, and reproduction of the Black-tailed Jack rabbit. *Journal of Mammalogy* 40: 63–81.
- Lloyd H. G. 1968. Observations on breeding in the brown hare (*Lepus europaeus*) during the first pregnancy of the season. *Journal of Zoology* 156: 521–528.
- Novaro A., Capurro A., Travaini A., Funes M. and Rabinovich J. 1992. Pellet-count sampling based on spatial distribution: a case study of the European hare in Patagonia. *Ecología Austral* 2: 11–18.
- Pépin D. 1989. Variation in survival of brown hare (*Lepus europaeus*) leverets from different farmland areas in the Paris basin. *Journal of Applied Ecology* 26: 13–23.
- Pielowski Z. 1969. Sex ratio and weight of hares in Poland. *Acta Theriologica* 19: 119–131.
- Raczyński J. 1964. Studies on the European hare. V. Reproduction. *Acta Theriologica* 9: 305–352.
- Suchentrunk F., Willing R. and Hartl G. B. 1991. On eye lens weight and other age criteria of the brown hare (*Lepus europaeus* Pallas, 1778). *Zeitschrift für Säugetierkunde* 56: 365–374.
- Tiemeier O. W. 1965. The Black-tailed Jack Rabbit in Kansas. 1. Bionomics. *Kansas Agricultural Experimental Station Technical Bulletin* 140: 1–37.
- Valentincic S. I. 1956. Resultate zweijähriger Beobachtungen und Studien über den idealen Zuwachs beim Feldhasen auf der Insel "Biserni otok". *Zeitschrift für Jagdwissenschaft* 2: 152–160.
- Watson J. S. 1957. Reproduction of the wild rabbit *Oryctolagus cuniculus* (L.) in Hawke's Bay, New Zealand. *New Zealand Journal of Science* 38: 451–482.

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