## Examination of Reproductive Performance of Roe Deer (Capreolus capreolus) in Hungary

## István Majzinger

University of Szeged, College of Agriculture, Department of Wildlife Management and Environmental Protection, Hódmezővásárhely mi@mfk.u-szeged.hu

#### SUMMARY

The objective of the research partly is to compare the reproduction performance of the populations living in different regions with regard to some special characteristics (age, condition).

When estimating the age through tooth wear and cementumlayer-counting there was a difference of 0.87 years in favour of the first one (r=0,840; p<0,001). I found cementum layers at 42% of the does in the study after examining the  $M_1$  teeth.

There was lose connection between the weight (eviscerated, with head and legs) and the KFI (r=0,296; p<0,01), and for further analysis, I used only the KFI as the index for condition.

The regional average KFI varied from 0.24-0.37 in fawns, 0.82-1.73 in does, with individual extremes of 0-4.05. Within the examined regions the highest index belonged to the prime-aged does, while the 1-year-olds had a lower rate, and it was the lowest in the does older than 8 years.

The rate of fertility was between 83,3(ns)-100% as we can see from the presence of the CL. All the examined does were fertile, except in one region, while among the female fawns in two regions I only found three with active ovaries. The average number of CL was 1.5-2.13, and this varied by regions; all in all it was the highest in the 2-7-year-old group (1.96) and in the ones over 8 years (2.00!), while it was lower in the does younger than 1 year (1.90). The high fertility of the does over 8 years is remarkable.

I could examine the number of embryos in two regions during the post-implantation period, and beside 100% fertility I found significant differences among the does, which can be associated with the condition. The ratio of CL carriers and the pregnant does was 100% and 73% in the two regions, the average number of CL were 1.92 and 1.72, while the average embryo number were 1.83 and 1.36 per doe. The difference between the CL and the embryo numbers on the two regions were 5% and 21%. The difference (prenatal loss) is in connection with the age (age class) of the doe. It is possible, however, that in some cases oestrus was not followed by gestation. But in roe deer, owing to the commonly known lack of luteolysis-mechanism (Flint et al., 1994), the regression of the CL of the does that did not get pregnant takes place in December and January, so the CL found in January cannot prove a previous pregnancy, which might have been followed by an abortion.

Although it has to be proven, it seems that the number of the CL (potential progeny) can be associated with the age (r=0,418; p<0,01) and the weight (r=0,312; p<0,01) of the doe, while the embryo number (realised progeny) is influenced by the age of the doe and probably by external factors.

It is essential to continue and extend the research to increase the reliability of the results and their correlation.

**Keywords:** reproduction, fertility, embryos, litter size, corpus luteum, implantation, kidney fat index, body weight

## **INTRODUCTION**

The roe deer numbers the largest among the spreading ruminant species in Europe, unintermittedly from the Mediterranian regions of Southern Spain to the subarctic part of Norway, from France to the Urals. Its particular flexibility is proven by the fact that the roe deer was able to adapt itself to the extreme conditions of this vast territory, on the one hand, with the adaptation of its morphological and physiological characteristics, and on the other hand with its individual and social behaviour. This species stands in the limelight of research in Western and Northern Europe, and a great number of rules, which seemed to be unquestionable in the past, have proved to be false concerning either the reproduction, the nutrition biology, or, for instance, space use. Due to the plasticity of the species, a typical feature characteristic of a region in a certain period is necessarily true elsewhere or another time. This is why it is essential to carry out long term research on small local populations repeatedly within certain periods or do it continuously.

The roe deer population in Hungary comprises more than three hundred thousand animals, and their number is still rising. Their significance in wildlife management cannot be questioned and yet only few research results were published in connection with the species during the past fifteen years.

It is important to know the reproduction performance of the roe deer and also the influencing factors in order to get to know the species better, on the one hand, and on the other to plan the harvest and control the population adequately. The reproduction biology of the roe deer is peculiar (embryonic diapause), certain processes are not clear even today (whether the roe deer is monooestrous or seasonally polyoestrous, the pre-implantation mechanism reactivates the dormant fetus), or the possibility of the does' influencing the sex of their young ones in the fetal and postnatal stages.

The following preliminary results were achieved during an examination planned for several years and carried out on different types of habitats on the Great Hungarian Plain<sup>\*</sup>, therefore, some results still have to be substantiated.

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# LITERATURE

The productivity of roe deer populations is high. As in most of the does older than a year of age, conception takes place at copulation, and the litter size per doe is also high, although there are significant differences within and among populations (*Table 1*).

Table 1

Reproduction indexes of roe deer based on national and foreign examinations

Source	Gestation (%)	Litter size (fawn/doe)
Bakkay et al. (1978)	94.4	2.04
Sugár (1979)	90.6	2.62
Kaluzinski (1982)	-	1.88
Fruzinski and Labudzki, 1982		
in: Csányi (1988)	-	1.82
Fodor (1983)	94.5	2.14
Farkas (1985)	87.0	1.82
Sempéré et al. (1989)	96.0	-
Hewison (1996)	-	1.92

One sure sign of gestation is the presence of the corpus luteum. If no conception takes place the corpus luteum gets quickly absorbed (Andersen, 1953 cit.: Faragó and Náhlik, 1997). The number of embryos is 96% of the corpora lutea (Strandgaard, 1972), so we can reliably estimate the number of the embryos on the basis of the corpora lutea. Conversely, Hermes et al. (2000) say that the presence of the corpora lutea prior to imlantation proves only the fact of the copulation and not the gestation. During the late copulation observed in roe deer (November-December) only the does which remained unfertilized in summer cycle for the first time (Fodor, 1983) or cycle again (Raesfeld, 1977). On the other hand Hermes et al. (2000) claim that the regression of the corpora lutea in does rutting but not fertilised takes place in December-January due to the lack of the luteolisis mechanism. Thus the roe deer is strictly monoestrous and the does which have not been fertilised when copulating are in anoestrous during the period of the embrionic diapause. At the same time there are reports on well-developed female fawns in good condition that occasionally some of them might copulate in November, but this is rarely followed by gestation and parturition. Sugár (1979) found gestation in three female fawns.

Potential litter size is determined by the body weight irrespective of the age of the doe (Hewison and Gaillard, 2001). In most ungulates body weight and age influence the reproductive performance and there can be a minimum age and body-weightthreshold for the first sussessful reproduction. At conception, body weight is important in the determination of the potential litter size, as it forms a kind of upper limit to the maximum reproductive performance, but the degree of the implantation of the blastocysts is influenced by age and external factors (weather condition) (Gaillard et al., 1992). Embrionic diapause makes it possible for the doe to control her reproductive input by selective implantation, which is a kind of answer to the seasonal changes of environmental conditions (Hewison and Gaillard, 2001). According to Farkas (1985) there is a definitely positive correlation between condition and litter size.

Implantation failure shows high diversity (16.7-54.5%) in adult does in a certain population, and it is much higher than the previously reported 8.3% and 9.4% (Borg, 1970 cit.: Hewison and Gaillard, 2001). At the same time neither the body-condition nor the body weight had an effect on implantation losses, but the possibility of the implantation of all blastocysts definitely depended on the age of the doe. A dramatic decrease of fecundity occurred over 7 years of age (Hewison and Gaillard, 2001).

# MATERIALS AND METHODS

We examined 128 roe deer (111 does and 17 female fawns) bagged between 1<sup>st</sup> October 2002 and 28<sup>th</sup> February 2003, the samples and data of which are from various types of habitats on the Great Hungarian Plain. Supposedly because of the different ecological conditions the biological parameters of the roe deer population living there and also the reproductive performance are different:

- Woody lowland I., II. forest 30%, meadows and pastures 20%, soil 40%, other 10%.
- Agricultural area and lowland forest 1%, meadows and pastures 17%, soil 75%, other 7%.
- Sandy soil agricultural area and woodland forest 27%, meadows and pastures 19%, soil 39%, vineyard and orchard 12%, other 3%.
- Intensive cultivation agricultural area I., II. forest 1%, meadows and pastures 5%, soil 83%, other 11%.
- *Plain and wood transitional area* forest 17%, meadows and pastures 11%, soil 46%, vineyard and orchard 11%, other 15%.

The age of the animals were estimated on the basis of tooth wear and of cementum layer counting; we measured the body weight after eviscerating (measured with head and legs). To indicate the condition I calculated kidney-fat index (KFI). To determine the fecundity and gestation parameters I examined the ova, the corpora lutea (CL), and the embryos (I determined their C-R length and also their sex.). Using these data I determined and compared the condition and the fecundity parameters on the regions concerned and I tried to find connections. In some cases the results are not reliable due to the low number of elements. I completed the MS Excel table programme.

# **RESULTS AND DISCUSSION**

# • Age estimation

Several characteristics, such as reproduction performance is in connection with the age of the

animals; this is why a reliable and correct age estimation has great importance. This happens on the basis of tooth wear in practice, which, however, has many possibilities for errors (i.e. lack of experience, individual genetic differences, the effect of habitat on the intensity of abrasion). Marosán (2001) says that the age of the roe deer, as with the red deer, can be estimated in Hungary with high accuracy by cementum layer counting, so this is the reason why I also chose this method as a control for tooth wear.

When estimating the age of the does, we got an average of 3.91 years, while we got 3.04 by cementum layer counting, so with the previous method we overestimated the age almost by one year (0.87). The correlation between the two method is r=0.840; p<0.001 (Figure 1), which proves the fact, that with adequate experience, the age of the roe deer can be estimated with great accuracy. After making a transversal cut and polishing the M<sub>I</sub> (molar) teeth from the jaws of the examined does I found a clearly visible layer on the cervical part of 42% of the samples. To improve the result, the examination should be repeated on the M<sub>II</sub> teeth with the above method, or on the horizontal polishing of the root (near the peak) of some of the incisor teeth (Marosán, 2001). Nevertheless to examine the reproduction performance and to explore its correlation with age, it is sufficient to put the animals into three age groups: 1 year, 2-7 years (prime-age) and over 8 years (old), and to do this we can trust and rely on the two methods mentioned above.



### • Body weight and condition

There was a weak correlation (r=0,269; p<0,01) between the eviscerated body weight (measured with head and legs) and the condition determined by the kidney-fat index [KFI = kidney fat (g)/kidney weight (g)] in the period from November to late February.

The condition of the does did not correlate closely with age (r=0,027; ns), but we can claim (*Figure 2*) that among the young and prime-aged animals there was a large dispersion concerning their condition, while in the old age group (in the game-bag), there were only animals with medium or weak conditions.

The extreme values of the KFI were 0.00-4.05 and the largest values were shown by the 2-7-yearold animals within the certain areas, while it was lower in the 1-year-old ones, and does over eight years had the lowest KFI. When examining it by regions: the body weight and the condition of the does showed significant differences, which presumably had an effect on mortality (*Table 2*). At the same time within the certain regions there was large dispersion especially at the KFI values.

Despite the extremely long and severe winter the roe deer population survived better than expected, especially in the regions where there had been intensive winter feeding. The values of their condition give an interesting picture in the examination period (between 5<sup>th</sup> November and 28<sup>th</sup> February), by regions. Although the average KFI of the does are different by regions; surprisingly, they generally did not decrease significantly (*Figure 3*).

When examining the values in each doe, the condition did not decrease tendentiously, but from January there was a significant dispersion, which might be a sign of competition among the does. It is possible that the does which could obtain more feed and had better protection (cover, shelter) were able to maintain or even improve their condition, while the fat reserve of the "losers" decreased considerably or was completely used up. The role of the body weight (competition?) seems to be proved by the fact that until mid-January, the body-weight-KFI correlation was r=0,181 (ns), from mid-January to late February it was r=0,471 (p<0,05), that is the role of the body weight increased in the maintenance of the condition.

We cannot say the same about the grown-up progeny, as the fawns were in comparatively good condition in December, but by the end of January and the beginning of February the severe winter had used up their fat reserves (*Figure 4*), because of their extreme sensitiveness.

Where the shooting took place in November and December, the feeding was intensive, and the animals were not disturbed in January and in February, there the prevention against high mortality was successful, mainly among the fawns.



Figure 2: Connection between age and KFI in does

		Bodywei	ght (kg)	Kidne		
Region	n	average	intervallum (P=0,05)	average	intervallum (P=0,05)	Mortality
Intensive agricultural I.	40	18.40	17.76-19.04	0.92	0.72-1.12	Medium (6-10%)
Intensive agricultural II.	13	16.16#*	14.94-17.38#*	1.13	0.74-1.52	Medium (6-10%)
Agricultural-lowland	25	18.61	17.92-19.28	1.25	1.07-1.45	Low (1-5%)
Woody lowland I.	4*	17.25	15.25-19.25*	0.92	0.17-1.67*	High (10%<)
Woody lowland II.	13	19.04	18.32-19.76	1.73	1.14-2.30	Minimal (<1%)
Sandy agr. and woody	12	17.92	16.93-18.91	0.82	0.53-1.11	nd
Transitional	6	19.41	17.11-21.71	0.98	0.46-1.50	nd

### Body weight, kidney fat index and mortality of does on different regions

The territorial differences between the body weight and the kidney fat index are significant (p<0,01).

\*: not significant; #: measured without head and legs; nd: no data



# Figure 3: Kidney fat index of the does

Figure 4: Kidney fat index of the fawns



## Fertility and the number of corpora lutea

I determined fecundity by comparing the number of does carrying CL to the number of bagged (shot) does and used the same method with female fawns (Table 3).

Except one region (Transitional), everywhere, each doe, regardless of age, carried at least one CL, which means they had a rut and could be fertile. The 83.3% result is not significant due to the low number of elements. From 17 female fawns in 3 regions there were only three animals which had active ovaries (at the end of January) with 1-2 diameter follicles but I found no CL.

There were significant differences among the regions in the number and distribution of the CL (Table 4, 5).

I could examine the relation of the number of CL, the age, the body weight and the KFI on one region, where the number of elements were sufficiently high (Intensive agricultural I.). There was no appraisable relation between the number of CL and the KFI (r=0,115; ns), it was weak between the number of CL and the body weight (r=0,312; p<0,1), while the correlation between the number of CL and the age of the doe was medial (r=0,418; p<0,01). It is noteworthy that the number of CL of the old does was the highest, which proves the fact that fertility in this age group did not deteriorate. The number of CL, however, cannot be identified with the number of prospective embryos, but the elderly does surely rut and the ovulation rate is not lower than that of the younger ones. The one-year-old group had the lowest CL number.

Table 3

Region	Fa	wn	1 y	ear	2-7 y	ears	Over 8	years	To (without	tal : fawns)
	n	%	n	%	n	%	n	%	n	%
Intensive agricultural I.	nd	-	8	100	27	100	4	100	39	100
Intensive agricultural II.	1	0	4	100	8	100	1	100	13	100
Agricultural lowland	nd	-	8	100	12	100	2	100	22	100
Woody lowland I.	9	0	nd	-	4	100	nd	-	4	100*
Woody lowland II.	7	0	nd	-	10	100	1	100	11	100
Sandy agr. and woody	nd	-	1	100	10	100	1	100	12	100
Transitional	nd	-	nd	-	4	75	2	100	6	83,3*
nd: no data										

Fertility by regions and age groups

\*: not significant

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Table 2

Feb

### Table 4

### The number of CL per does on different regions

Region	0	1	2	3	Average number
		OF CL			
Intensive agricultural I.	0.0	0.0	92.3	7.7	2.10
Intensive agricultural II.	0.0	17.0	83.0	0.0	1.84
Agricultural lowland	0.0	0.0	86.4	13.6	2.13
Woody lowland I.	0.0	0.0	100.0*	0.0	2.00*
Woody lowland II.	0.0	7.0	92.2	0.8*	1.91
Sandy agr. and woody	0.0	25.0	75.0	0.0	1.75
Transitional	16.0*	17.0*	67.0*	0.0	1.50*
Average	1.0*	7.7	84.5	6.8	1.98

The differences among the average corpus lutea numbers on different territories are significant (p<0,01).

\*: not significant

Average number of CL by regions and age groups

Table 5

Region	1 year	2-7 years	Over 8 years	Total, without fawns
Intensive agricultural I.	2.00	2.10	2.33	2.10
Intensive agricultural II.	1.50	2.00	2.00*	1.84
Agricultural lowland	2.00	2.21	2.00*	2.13
Woody lowland I.	nd	2.00	nd	2.00*
Woody lowland II.	nd	1.88	2.00*	1.91
Sandy agr. and woody	2.00*	1.70	2.00*	1.75
Transitional	nd	1.50	1.50*	1.50*
Average	1.90	1.96	2.00	1.98

nd: no data

\*: not significant

## • Post implantation examination

The reactivation (expansion, elongation) and implantation of the dormant blastocysts take place in late December and January. The question arises whether there is a change in quantity or quality (sex ratio) in the fecundated ovules compared to the implanted ones during the five months of the embryonic diapause. This long period might be an opportunity for the does to optimise their reproductive inputs according to their own physical condition and external environmental influences, that is with the possible decrease of the litter size (total number of blastocysts) to maximise the quantity of the realised (implanted, born) progeny, in order to achieve long-term reproductive success.

I could examine the post-implantation conditions on two regions (*Woody lowland II., Sandy agr. and woody*) with a limited number of elements (n=23).

The differences between the number of CL and the embryos were 5% and 21% in the two territories, the latter where the average body weight was lower and the condition was weaker (*Table 6*). The question is still to be answered whether the embryos of the does carrying CL but not pregnant were absorbed or whether these does (or at least some of them) had not been pregnant at all.

To decide this, in the future I plan to examine the quantitative pre-implantation conditions of the CL and the blastocysts.

The ratio of the quantity of the CL and the embryos in the case of the one-year-old and the over-8-year age groups is still to be examined because of the low number of elements. Nevertheless, the differences between the numbers of embryos and CL is 29% in the one-year-old, 10% in the prime-aged and 14% in the over-8-year old (*Table 7*).

In the does where the number of CL was different from the number of embryos, there were no embryos at all in 70% of the cases, that is the principle of "all or nothing" phenomenon (Short and Hay, 1966), which means they lost all their embryos or none (if we consider it prenatal loss).

All in all, it seems that the number of CL (potential progeny) can be related mainly to the age (r=0,418; p<0,01) and the body weight (r=0,312; p<0,1) of the doe, while the number of the embryos (realised progeny) is influenced by the age (age group) of the does, their body weight (r=0,480; p<0,05) and perhaps by external factors as well.

Embryonic development becomes more intensive from late January (*Figure 5*), but the single embryos are not larger than any of the twin-embryos, at least not in this period. Presumably, in this early stage of the gestation the feeding of the twin-embryos does not mean a greater task for the doe than the feeding of singletons. The measurements (CR-length) of the twin embryos (within one uterus) were nearly the same (r=0.998; p<0.01).

Table 6

Table 7

Post im	plantation	results	in	two	regions
I Obt mit	pitation	1 Courto	***		regions

Region	n	Average body weight kg	Average KFI	Fertility %	Average number of CL	Average number of embryos	Embryo/ CL ratio
Woody lowland II.	12	19.04	1.73	100	1.92	1.83	0.95
Sandy agr. and woody	11	18.00	0.79	100	1.72	1.36	0.79

#### CL and embryo number by age groups after implantation

	1 year n=4	2-7 years n=22	Over 8 years n=4	Total n=30
CL/doe	1.75	1.86	1.75	1.83
Embryo/doe	1.25	1.68	1.50	1.60

#### Figure 5: Changes in embryo measurements



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