Influence of inbreeding on the incidence of cryptorchidism in males of Lowland-line bison

ELŻBIETA CZYKIER, JERZY DACKIEWICZ*, MICHAŁ KRZYSIAK*,
MATEUSZ CHOLEWSKI**, WANDA OLECH***

Department of Histology and Embryology, Medical University of Białystok, Waszyngtona 13, 15-269 Białystok, Poland

*Białowieża National Park, Białowieża, Poland

**Student Research Society at the Department of Histology and Embryology, Medical University of Białystok, Poland

***Department of Genetics and Animal Breeding, Warsaw University of Life Sciences,
Ciszewskiego 8, 02-786 Warsaw, Poland

Received 06.08.2015

Accepted 04.03.2016

Czykier E., Dackiewicz J., Krzysiak M., Cholewski M., Olech W.

Influence of inbreeding on the incidence of cryptorchidism in males of Lowland-line bison

Summary

The aim of this study was to find whether the coefficient of inbreeding is correlated with the incidence of cryptorchidism in the Białowieża line of bison.

Our study included 605 male European bison *Bison bonasus* (L.) born between 1971 and 2015 in the Białowieża Forest. The animals were free-ranging as well as from captive breeding programs. Among the animals culled, there were 144 European bison calves. The calves were not considered in the research, because the descending of the testes in European bison may not occur until a year after birth. The study period, from 1966 to 2015, was divided into 5 decades. The number of male European bison born in each decade was recorded, as well as the number of cryptorchidism cases identified through necropsy in these animals. The percentage of incidence of cryptorchidism among them was calculated for every decade. Subsequently, the Pearson correlation coefficient was calculated between the number of cryptorchidism cases and the coefficient of inbreeding.

In 461 male European bison older than 1 year, there were 18 cases of cryptorchidism. In the animals born in the first decade, two autopsical cases of cryptorchidism (0.045%) were recorded, and 4 cases of cryptorchidism (0.03%) were found in the animals from the second decade. Among the bison born in the third decade, there were no cases of cryptorchidism. In the fourth decade, 7 cases of cryptorchidism (0.071%) were diagnosed, and among the bison born during the fifth decade, there were 5 cases (0.111%).

The Pearson correlation coefficient suggests that, as the value of the coefficient of inbreeding rises, there is an increase in cryptorchidism cases in male European bison.

Keywords: *Bison bonasus*, cryptorchidism, inbreeding

The reintroduction of the European bison *Bison bonasus* (L.) was proposed in 1924 along with the establishment of the International Society for the Protection of the European Bison. At that time, there were 54 European bison in zoos and nature preserves. However, after genetic testing, it became apparent that the Lowland (Białowieża) bison descends from only seven individuals (18-20, 23). Therefore, all animals are very closely related. According to Bielousova (3), the population of the European bison recreated in the Białowieża Forest possesses only 5 sets of diploid genes. Research conducted by Olech (17, 21) shows that the coefficient of inbreeding increases in successive generations of the European bison.

The literature concerning cryptorchidism in the Białowieża line of male European bison is very limited. Koziorowski et al. (14) mention cryptorchidism

as one of the abnormalities observed in the reproductive system of the European bison. Other authors (7, 15, 25) list cases of cryptorchidism in European bison, specifying whether it is unilateral or bilateral. Czykier (4), on the other hand, presents a histological description of a European bison testis that did not descend into the scrotum, compared to one that did. The abovementioned authors do not, however, examine the correlation between the coefficient of inbreeding and the frequency of cryptorchidism in the Białowieża line of the European bison.

Research conducted on other species of animals proves that inbreeding leads to a series of developmental defects, including cryptorchidism. Hence the decision to assess the correlation between the coefficient of inbreeding and the incidence of cryptorchidism in the Białowieża line of bison.

Material and methods

Since 1971, during autumn and winter months, a selective cull of the European bison is conducted in the Białowieża Forest. The causes include disease, especially posthitis/balanoposthitis in males, injuries, as well as poor physical condition and age (16). Selective culls are carried out during early morning hours. Dissections of the animals killed are conducted according to an established protocol, in which macroscopic and microscopic pathomorphological changes are recorded. The study included 605 male European bison born between 1971 and 2015 in the Białowieża Forest. The animals were free-ranging as well as from captive breeding programs. The animals culled included 144 calves. These, however, were not considered in the research, since, according to Swieżyński (25), the testes in European bison may not descend until one year after birth. The study period, from 1966 to 2015, was divided into 5 decades. The numbers of male European bison born in each decade were recorded, as well as the number of cryptorchidism cases identified through necropsy in these animals. The percentage of incidence of cryptorchidism among male European bison was calculated for each decade. Subsequently, the Pearson correlation coefficient was calculated between the number of cryptorchidism cases and the coefficient of inbreeding.

Results and discussion

Among 461 male European bison older than 1 year, there were 18 cases of cryptorchidism, including 2 cases of bilateral cryptorchidism (one animal was 13.5 and the other 3 years old) and 16 cases of unilateral cryptorchidism, right-sided or left-sided (in animals aged 1.5 to 12 years) (Tab. 1). The diagnosis of cryptorchidism in male European bison at such an advanced age (as old as 13.5 years) is due to the fact that the recognition of cryptorchidism in this species is possible only post-mortem. It is impossible to determine whether both testes are within the scrotal sac in a live individual.

In male European bison born in the first decade, two autopsical cases of cryptorchidism (0.045%) were recorded, and 4 cases of cryptorchidism (0.03%) were found in animals from the second decade. Among bison born in the third decade, there were no cases of cryptorchidism. In the fourth decade, there were 7 cryptorchidism cases (0.071%), whereas among bison born during the fifth decade, there were 5 cases (0.111%) (Tab. 1).

The Pearson correlation coefficient suggests that as the value of the coefficient of inbreeding rises, there is an increase in cryptorchidism cases in male European bison (Fig. 1).

The descent of testes, or the migration of the male gonads from the posterior wall of the abdominal cavity to the scrotal sac, occurs during the fetal stage. The testes, located outside the peritoneum, migrate into the deep inguinal rings, and then move into the superficial inguinal rings to finally reach the scrotal sack. Inside the scrotum, the testes move to the lower end of the

Tab. 1. Decades, number of calves born in 1966-2015, number of cryptorchidism cases, percentage of cryptorchidism, coefficient of inbreeding

Decade	Number of calves born	Cryptorchidism		Coefficient of inbreeding
		Number of cases	%	
1 1966-1975	44	2	0.045	0.323
2 1976-1985	135	4	0.030	0.358
3 1986-1995	139	0	0.000	0.415
4 1996-2005	98	7	0.071	0.409
5 2006-2015	45	5	0.111	0.480
Altogether	461	18	0.039	0.397

Fig. 1. Correlation between coefficient of inbreeding and percentage of cryptorchidism in male European bison

Explanation: 1 – 1st decade from 1966 to 1975; 2 – 2nd decade from 1976 to 1985; 3 – 3rd decade from 1986 to 1995; 4 – 4th decade from 1996 to 2005; 5 – 5th decade from 2006 to 2015

spermatic cord (2, 10, 13). The descent of the testes in different species takes place at various stages of development. In pigs, horses, cattle, and sheep, it is completed before birth, whereas in bison, rabbits and dogs, it continues in the postnatal period (9, 25, 28, 29).

Cryptorchidism, a condition in which testes do not descend into the scrotum, occurs with various frequency for different species of animals. For goats (bucks), the frequency of occurrence is 1.1%, in Andean deer 2.9%, in rams 5.5%, in male rabbits 1.8%, and in male horses $\leq 1\%$ (2, 6, 12, 22, 26, 27). Undescended testes may be found within the abdominal cavity, in the inguinal canal, in the superficial inguinal ring or at the base of the penis. Most often they are found within the inguinal canal (5, 10). The descent of the testes, as described by Hutson (10), can be divided into three phases: abdominal, inguinal, and scrotal. A different opinion is expressed by Klonish (13), who believes that testicular descent occurs in two phases: intra-abdominal and inguinal-scrotal. All authors, however, agree that the descent of the testes is a complex process influenced by genetic, environmental and hormonal factors (10, 13).

According to Hutson (11), in the intra-abdominal phase of the descent, a key role is played by the gubernaculum, which directs the migration of the gonads from the posterior abdominal wall toward the inguinal canal. The normal development of the gubernaculum is

controlled by insulin-like hormone 3 (INSL3) produced by fetal Leydig cells, or transformed steroidogenic cells (1, 8, 24). At the same time, within the somatic cells of the developing gonads, the activation of the *Sry* gene occurs, which causes the cells to transform into supporting cells. The supporting cells begin to produce Müllerian-inhibiting substance (MIS), also known as anti-Müllerian hormone (AMH), which inhibits, by the way of apoptosis, the development of the Müllerian ducts (2, 10, 13).

The inguinal phase of testes descent, on the other hand, occurs mainly under the control of testosterone produced by the fetal-type Leydig cells.

Cryptorchidism can be unilateral or bilateral (30). Unilateral cryptorchidism in animals occurs significantly more often than bilateral cryptorchidism (1). The left cryptorchid testis in dogs is usually located in the abdominal cavity, while the right cryptorchid testis can be found in the inguinal canal (1). The cases of cryptorchidism in European bison recorded in our study deviated from this rule, since both left and right cryptorchid testes were found in the inguinal canal.

The results of our research show that in the Białowieża line of European bison the occurrence of cryptorchidism is correlated with the coefficient of inbreeding. Since cryptorchidism is a developmental defect, it could be presumed that in future generations of the European bison it will occur more frequently than it has until now.

References

- Almeida J., Conley A. J., Ball B. A.: Expression of anti-Müllerian hormone, CDKN1B, connexin 43, androgen receptor and steroidogenic enzymes in the equine cryptorchid testis. *Equine Vet. J.* 2013, 45, 538-545.
- Amann R. P., Veeramachaneni D. N. R.: Cryptorchidism in common eutherian mammals. *Reproduction* 2007, 133, 541-561.
- Bielousova I. P.: Znachenie i ocenka pokazatelej geneticheskogo raznoobrazija dlja reshenija problemy sokhraneniya evropeiskogo zubra. Prirodozashchita i razvitiye biosfernyj zapovednik. Pushchino 1999, p. 1-108.
- Czykier E.: The case of left cryptorchid testis in the European bison – histological and morphometric description. *Med. Weter.* 2011, 67, 774-777.
- Felumlee A. E., Reichle J. K., Hecht S., Penninck D., Zekas L., Yeager A. D., Goggin J. M., Lowry J.: Use of ultrasound to locate retained testes in dogs and cats. *Vet. Radiol. Ultrasound*. 2012, 53, 581-585.
- Flueck W. T., Smith-Flueck J. A. M.: Age-independent osteopathology in skeletons of a South American cervid, the Patagonian huemul (*Hippocamelus bisulcus*). *J. Wildl. Dis.* 2008, 44, 636-648.
- Gill J.: Zarys fizjologii zubra. Wydawnictwo Severus, Warszawa 1999, p. 42-60.
- Giesecke K., Sieme H., Distl O.: Infertility and candidate gene markers for fertility in stallions: a review. *Vet. J.* 2010, 185, 265-271.
- Hayes Jr H. M.: Epidemiological features of 5009 cases of equine cryptorchidism. *Equine Vet. J.* 1986, 18, 467-471.
- Hutson J. M., Hasthorpe S., Heyns C. F.: Anatomical and functional aspects of testicular descent and cryptorchidism. *Endocr. Rev.* 1997, 18, 259-280.
- Hutson J. M., Hasthorpe S.: Testicular descent and cryptorchidism: the state of the art in 2004. *J. Pediatr. Surg.* 2005, 40, 97-302.
- Kafi M., Oryan A., Morgan-Azghadi N.: Pathology of testis and epididymis in native goats in southern Iran. *Comp. Clin. Pathol.* 2007, 16, 201-205.
- Klonisch T., Fowier P. A., Hombach-Klonisch S.: Molecular and genetic regulation of testis descent and external genitalia development. *Dev. Biol.* 2004, 270, 1-18.
- Koziorowski M., Seremak B., Giżejowski Z., Gilu P., Kozioł K., Kowal E., Jagustyn B., Sulik M., Szeleszczuk O., Olech W., Söderquist L., Glogowski J.: Season controlled reproduction of undomesticated animals. *Reprod. Biol.* 2006, 6, 137-149.
- Krasińska M., Krasiński Z. A.: European bison. The Nature Monograph. Mammal Research Institute, Polish Academy of Sciences, Białowieża 2007, p. 1-317.
- Krasińska M., Krasiński Z. A.: Przebieg i dyspersja choroby nekrotycznego zapalenia napletka samców zubra na terenie polskiej części Puszczy Białowiejskiej. *Parki Nar. Rez. Przyr.* 2010, 29, 107-128.
- Olech W.: Analysis of inbreeding in European bison. *Acta Theriol.* 1987, 32, 373-387.
- Olech W.: Ocena zmienności populacji zubrów na podstawie rodowodu, [in:] Kita J., Anusz K. (ed.): Health threats for the European bison, particularly in free-roaming populations in Poland. The SGGW Publishers, Warszawa 2006, p. 211-215.
- Olech W.: Perspektywy restytucji zubrów w Europie, [in:] Olech W. (ed.): Perspektywy rozwoju populacji zubrów. Wydawnictwo ARTISCO Goczałkowice-Zdrój 2006, p. 37-42.
- Olech W.: The number of ancestors and their contribution to European bison (*Bison bonasus* L.) population, [in:] Kita J., Anusz K. (ed.): Health threats for the European bison, particularly in free-roaming populations in Poland The SGGW Publishers, Warszawa 2006, p. 229-236.
- Olech W.: Wpływ inbrodu osobniczego i inbrodu matki na przeżywalność cieląt zubra (*Bison bonasus* L.). *Rozprawy Naukowe i Monografie. Wydawnictwo SGGW*, Warszawa 2003, p. 1-86.
- Regassa F., Terfe F., Bekana M.: Abnormalities of the testes and epididymis in bucks and rams slaughtered at Debre Zeit abattoir, Ethiopia. *Trop. Anim. Health. Pro.* 2003, 35, 541-549.
- Statis H. M.: An analysis of inbreeding in the European bison. *Genetics* 1960, 45, 275-287.
- Stout T. A. E.: Can and should we do more to reduce the incidence of cryptorchidism? *Equine Vet. J.* 2013, 45, 531-532.
- Świeżyński K.: The male reproduction organs of the European bison. *Acta Theriol.* 1968, 13, 511-551.
- Tarigan S., Ladds P. W., Foster R. A.: Genital pathology of feral male goats. *Aust. Vet. J.* 1990, 67, 286-290.
- Watt D. A.: Testicular pathology of merino rams. *Aust. Vet. J.* 1978, 54, 473-478.
- Wensing C. J. G.: Testicular descent in some domestic mammals: I, Anatomical aspect of testicular descent. *Proc. K. Ned. Akad. Wet. Ser. C.* 1968, 71, 423-434.
- Wensing C. J. G.: Testicular descent in some domestic mammals: II, The nature of the gubernacular change during the process of testicular descent in the pig. *Proc. K. Ned. Akad. Wet. Ser. C.* 1968, 76, 190-195.
- Wright S.: Testosterone concentrations in cryptorchids. *Equine Vet. Educ.* 2013, 25, 599-600.

Corresponding author: Assoc. Prof. Elżbieta Czykier, MD, PhD, Waszyngtona 13, 15-269 Białystok, Poland; e-mail: elzbieta.czykier@umb.edu.pl