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An Analysis of the Genetic Relationships and Inbreeding in Tatra Shepherd Dogs Depending on the Type of Breeding

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Original article

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The aim of the study was the analysis of the structure of the population, relationship coefficients, and inbreeding trends in terms of: sex, breeding system, and inbreeding degree of Tatra Shepherd dogs. The breed's inbreeding rate was 6.34%, and for a standardised 4-generation population was 6.68%. The highest inbreeding rate was found in "non-champion-dogs" and in "Polish dog" groups consisting of males and females. The limit value F_X was exceeded for 25.65% of Tatra dogs, and the critical value was exceeded for 11.52%. An increasing ancestor loss coefficient (AVK) was found, which may result in an increased number of inbred animals. In particular, this referred to female dogs in the nCH, PL, and Z groups, whereas a significant increase of AVK was observed in the group of male dogs from foreign kennels. The resulting COR values were 55.58% for males and 55.44% for females.

Key words: Tatra Shepherd Dog, pedigree analysis, inbreeding value, ancestor loss.

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There are about 500 million dogs around the world, including more than 63 million living in the EU. The highest number of dogs in Europe live in Russia – more than 15 million. In Germany there are 9,4 million, 9 million in the United Kingdom, and 7,6 million in Poland. In Poland and Romania, it is estimated that 42% of households have at least one dog, which puts Poland first in Europe. The World Canine Organization (Fédération Cynologique Internationale, FCI) has classified 344 breeds, and Poland has currently 340 registered breeds split into hunting, guardian, and shepherd dogs (YOUNG *et al*. 2011; HUMANE SOCIETY US 2016; FCI 2017; FEDIAF 2018; HOROSZEWICZ *et al*. 2017). Recently, dog shows have become increasingly popular, and selection has been more oriented towards the phenotype and grooming of dogs (LINDBLAD-TOH *et al.* 2005). Studies show that such breeding leads to a loss of genetic variability in some breeds in Europe and in the world (LEROY *et al.* 2006; CALBOLI *et al.* 2008; VOGES & DISTL 2009; MORTLOCK *et al*. 2016; KETTUNEN *et al*. 2017; MOSTERT *et al*. 2015; JANSSON & LAIKRE 2018).

The oldest canine organization in Poland is the Polish Kennel Club (ZKwP – founded in 1938) which keeps Stud Books (KW) for Polish breeds and maintains the longest pedigree lines for all world recognized breeds, verifies pairs of parents and registered litters, conducts mental health tests, and organizes shows of purebred dogs. ZKwP is a patron of 5 national breeds including: the Polish Grey Hound (FCI standard no. 333), the Polish Hunting Dog (FCI standard no. 354), the Polish Hound (FCI standard no. 52), the Polish Lowland Sheepdog (FCI standard no. 251), and the Tatra Shepherd Dog (FCI stanadard no. 252). The first pedigree Tatras in Poland were born in 1957 in Łeba, in Danuta's Hryniewicz kennel and started the postwar history of the breed (REDLICKA & REDLICKI 2003; POLISH KENNEL CLUB 2009).

Despite their unique guarding efficiency, Tatras indirectly contribute to the green grazing of sheep

(RADZIK-RANT & WOJNARSKA 2008; CHODZEŃ 2014; FCI 2017). The main features that predispose them to this type of work are endurance, strength, courage, distrust of strangers, resistance to various weather conditions, good hearing and sight, and a well-developed defensive instinct (ARHANT *et al*. 2010; HAVERBEKE *et al*. 2010).

Despite many genetically valuable Tatra Shepherd dogs having been taken abroad from Poland by their owners, the population seems to be reconstructed and quite even. However, it is still not sufficiently numerous, which prompts the monitoring of relationships in order to avoid inbreeding (KALINOWSKA *et al.* 2010; KANIA-GIERDZIEWICZ *et al*. 2015).

Inbreeding increases the level of homozygosity of an animal, which has positive and negative effects. Creating homozygous animals can reveal latent defects in recessive genes. Many authors claim that the loss of genetic variability and inbreeding contributes to the development of physical diseases, defects, and disorders such as reduced fertility and prolificacy, the occurrence of lethal alleles in a litter, and a lower offspring survival rate. In addition, anomalies may occur in the hip joint anatomy leading to dysplasia among house dogs, especially in large breeds (HALL & WALLACE 1996; UBBINK *et al*. 1998; ÓLAFSDÓTTIR & KRISTJÁNSSON 2008; KRZEMIŃSKA *et al.* 2018). Inbreeding coefficients for a breed range from about 0.82% for Golden Retrievers, 2% for German Shepherd Dogs, 4.5% for Bullmastiff Dogs, 6.7% for Bracco Italiano Dogs, through 9% for Great Danes to 26% for Nova Scotia Duck Tolling Retrievers and as much as 37% for Polish Hounds (DROZD & KARPIŃSKI 1997; GŁAŻEWSKA 2008; MÄKI 2010; KANIA-GIERDZIEWICZ *et al.* 2014; CECCHI *et al.* 2016; MORTLOCK *et al.* 2016). The F_x coefficient was supplemented with by information about the content of a unique pool of genes revealed by the Ancestor Loss Coefficient (AVK). The AVK makes it possible to assess inbreeding in previous generations and indicates a percentage ratio of unique ancestors to the total number of ancestors for a specific number of generations (SCHLEGER & STUR 1990; CALBOLI *et al.* 2008; JANSSON & LAIKRE 2014; KRUZIŃSKA *et al.* 2019).

The DNA polymorphism analysis demonstrated a considerable genetic differentiation of microsatellite markers and a lack of inbreeding in the analysed population of Tatra Shepherd dogs. Other studies conducted in Poland (KALINOWSKA *et al*. 2010; KANIA-GIERDZIEWICZ & GIERDZIEWICZ 2015) confirm the differentiated level of inbreeding with an upward trend, depending on the sex and age as well as the region of occurrence of the Tatra Shepherd Dog breed.

The studies were focused on an analysis of the structure of the population, estimating the relationship and inbreeding trend based on pedigrees taking into account factors such as: sex, breeding type (champions, non-champions), and origin (domestic and foreign) of Tatra Shepherd dogs. The main aim of the research was to determine the level of inbreeding in the breed and to deduce how it affects the breed maintenance, as inbreeding is inevitable due to a small breed population.

Material and Methods

Pedigree analysis allowed for the identification of entire pedigree lines thanks to the use of Pedigree Publisher by Breedmate software. The response included dogs with unknown ancestors, exterior corresponding to the breed standard, as well as dogs whose origin has been established up to 18 generations. The number of funders for the analyzed population was 98 animals. The relationship coefficients were analyzed among all 491 animals i.e. all 121,746 possible pairs were calculated according to the recursive by TIER (1990) with an algorithm modified GIERDZIEWICZ & KANIA-GIERDZIEWICZ (2007). A pedigree database containing the data of 491 Tatra Shepherds (198 male dogs and 293 female dogs) born between 1964-2014 was developed and used. To get a full picture of the changes in the inbreeding value of this breed, two periods of about ten years each were selected: 1994-2004 and 2005-2014. The Wright coefficients of inbreeding (F_x) and the ancestor loss coefficient (AVK) were estimated for all animals split according to: sex, having a champion title (CH), or not having a champion title (nCH), origin: from Polish kennels (PL) and from foreign kennels (Z). In addition, the coefficient of the relationship between the proband and its direct (R_{XA}) and collateral relationship (R_{XY}) was estimated.

The inbreeding coefficient of relationship (R_{XA}) and collateral relationship (R_{XY}) is calculated according to the following formulas (KANIA-GIERDZIEWICZ 2008; WRIGHT 1922):

Inbreeding coefficient:

$$F_X = \sum\left(\frac{1}{2}\right)^{n_1+n_2}\left(1+F_A\right)$$

Collateral relationship:

$$R_{XY} = \frac{\sum\left(\frac{1}{2}\right)^{n_1+n_2}\left(1+F_A\right)}{\sqrt{\left(1+F_X\right)\left(1+F_Y\right)}}$$

Relationship:

$$R_{XA} = \sum\left(\frac{1}{2}\right)^{n}\sqrt{\frac{\left(1+F_A\right)}{\left(1+F_X\right)}}$$

Key to formulas:

F_X – inbreeding coefficient of specimen $_X$; F_Y – inbreeding coefficient of specimen $_Y$; F_A – inbreeding coefficient of a common ancestor $_A$; R_{XA} – coefficient of kinship between specimen $_X$ and its ancestor $_A$; R_{XY} – coefficient of kinship between specimens $_X$ and $_Y$; n_1, n_2 – number of paths from the parents of specimen X to an ancestor A shared by the parents.

The calculations were made using Pedigree Explorer software (Wild Systems P/L, Australia). Collateral relationship was calculated with the support of the CFC program (SARGOLZAEI *et al.* 2006). The results of the study were subjected to an analysis of variance taking the following features into account in several different models: sex (male, female); champion (CH), non-champion nCH), domestic kennel (PL), and foreign kennel (Z). The data are presented as means with their standard deviations (±SD). The significance of differences between the mean values for respective groups was verified by the Tukey's test ($p<0.05$ and $p<0.001$) using Statistica 12 software (STATSOFT Inc. 2016).

Table 1

Average inbreeding value

Index	Sex	The whole population[1]	
		n	Mean± SD
F_x Total	Male Dogs	198	3.14 ± 0.46
	Female Dogs	293	2.79 ± 0.42
	Total	491	2.93 ± 0.44
F_x Inbred animals	Male Dogs	99	6.29 ± 0.47
	Female Dogs	128	6.38 ± 0.42
	Total	227	6.34 ± 0.44

F_x – Inbreeding value expressed in %,
[1]The whole population – 1-18 generational pedigree.

Results

Since 2003, a year-on-year increase has been observed in the number of Tatra Shepherd dogs newly registered with the divisions of the Polish Kennel Club. Every year, more females than male dogs are registered. As of December 31, 2016, out of 587 registered dogs, as many as 246 were qualified for breeding, which corresponded to 41.9% of all the registered animals of this breed. Over only five years (2011-2016), 1,961 puppies were born from 337 litters, which accounts for an average of about 5.8 puppies per litter. In 2016, there were 121 registered Tatra Shepherd dog breeders in Poland.

Among the 491 animals considered in the study, 227 individuals (46.23%) were inbred (Table 1). The average inbreeding value, F_X, throughout the analyzed population from the stud book (KW) and with 1 to 18-generation pedigrees was 2.93 ± 0.44%. Only for inbred animals F_X amounted to 6.34 ± 0.44%. The average values of F_X were 3.14% and 2.79%, respectively, for all male dogs and female dogs, and 6.29%, 6.38% for inbred male dogs and inbred female.

For the purposes of further calculations, pedigrees were divided into: champions (CH), non-champions (nCH), and dogs bred in Poland (PL) and abroad (Z) (Table 2). The average inbreeding value for Tatra shepherd dogs with full, 4-generation pedigrees, born in 1994-2014 amounted to 6.52 ± 0.48% for males and 6.79 ± 0.44% for females. The highest inbreeding rate was observed among males (7.08 ± 0.50%) and females (7.12 ± 0.45%) in the group of non-champions (nCH) and among males (6.75 ± 0.46%) and females (6.91 ± 0.44%) born in Polish kennels (PL). The level of inbreeding among Tatra Shepherd dogs born in 1994-2004 was 5.87 ± 0.50% for males and 4.88 ± 0.46% for females, whereas in 2005-2014 it was 6.94 ± 0.47% and 8.22 ± 0.42%, respectively. The lowest inbreeding value

Table 2

Average inbreeding value in the population of Tatra Shepherd dogs, taking into account their sex, group, and the studied years

Analyzed years	Male Dogs					Female Dogs				
	Total	CH	nCH	PL	Z	Total	CH	nCH	PL	Z
	n=82	n=31	n=51	n=71	n=11	n=112	n=28	n=84	n=96	n=16
$F_x$mean (1994-2004)	5.87	6.04	6.62	6.22	4.33	4.88	5.38	4.60	5.08	3.88
±SD	0.50	0.41	0.51	0,48	0.61	0.46	0.40	0.46	0.46	0.38
$F_x$mean (2005-2014)	6.94	4.90	7.58	7.06	5.86	8.22	6.46	8.58	8.19	8.40
±SD	0.47	0.41	0.50	0.45	0.49	0.42	0.38	0.42	0.43	0.37
$F_x$mean (1994-2014)	6.52	5.60	7.08	6.75	5.02	6.79	5.80	7.12	6.91	6.14
±SD	0.48	0.41	0.50	0.46	0.58	0.44	0.39	0.45	0.44	0.37

SD – Standard deviation, F_x – Inbreeding value expressed in %, CH – Champion, nCH – Untitled champion, PL – Bred in Poland, Z – Bred abroad.

(3.88 ± 0.38%) was recorded for female dogs born in 1994-2004 outside Poland, and the highest (5.38%) in the CH group. In the last analyzed group (2005-2014) the highest inbreeding value was found in the nCH (8.58%), compared to the lowest value for the CH group (6.46%). In the years 1994-2014 the lowest inbreeding value was found in females from the CH group (5.80%), compared to the nCH, PL, and Z groups. In male dogs the lowest inbreeding value was found in group Z (4.33%) analyzed in the years 1994-2004. Similarly, for group Z in relation to CH, nCH, and PL, the lowest inbreeding value was 5.02% in the analyzed years 1994-2014. A decreased value of inbreeding among male dogs born in 2005-2014 in comparison to dogs born in 1994-2004 was observed only in the group of champion dogs.

Inbreeding values (F_X) among Tatra Shepherd dogs showed the lowest number of specimens in the class of non-related animals and in particular male dogs (Table 3). The highest number of animals, i.e. 54.97% was reached in the group with Fx values from 1.5% to 6.5%. In this group there were 270 individuals (101 males and 169 females), whereas in the non-inbred group there were only 14 animals (3 male and 11 female). The limit value is 6.5% and the critical value is 12.5% (FALCONER 1996). The analysis shows that 126 of the Tatra Shepherd dogs, 47 males and 79 females, exceeded the limit inbreeding value. Furthermore, 11.52%, 56 animals (respectively 12.50% 25 males and 10.81% 31 females) exceeded the inbreeding value of 12.5%.

The average ancestor loss coefficient (AVK) within the entire analysed population (Table 4) entered in the stud book (KW) and with 1 to 18-generation pedigrees amounted to 92.30 ± 9.85%, including 84.81 ± 9.53% for inbred animals. However, for a standardized population it is 82.61 ± 9.75%, including 82.31 ± 9.65% for inbred animals.

Table 3

Range of inbreeding value for Tatra Shepherd dogs

Range	Total	Male Dogs	Female Dogs
F_x=0%	14	3	11
0%<F_x≤1.5%	25	22	3
1.5%<F_x≤6.5%	270	101	169
6.5%<F_x≤12.5%	126	47	79
12.5%<F_x	56	25	31

Data F_x represents the mean percentage share in the group.

Table 4

The average ancestor loss coefficient (AVK) in the population of Tatra Shepherds dogs (mean ± SD).

Index	Sex	The whole population[1]	
		n	Mean ± SD
AVK Total	Male Dogs	198	91.13 ± 9.88
	Female Dogs	293	92.42 ± 9.85
	Total	491	92.30 ± 9.85
AVK Inbred animals	Male Dogs	99	85.12 ± 9.44
	Female Dogs	128	84.57 ± 9.64
	Total	227	84.81± 9.53

[1]The whole population – 1-18 generational pedigree.

The values of the ancestor loss coefficient for Tatra Shepherd dogs pedigrees split into groups: CH, nCH, PL, and Z are presented in Table 5. The highest AVK was noted for male dogs (84.30 ± 9.09%) and female

Table 5

Values of the ancestor loss coefficient (AVK) in the population of Tatra Shepherd dogs, taking into account their sex, group, and the studied years

Analyzed years	Male Dogs					Female Dogs				
	Total	CH	nCH	PL	Z	Total	CH	nCH	PL	Z
	n=82	n=31	n=51	n=71	n=11	n=112	n=28	n=84	n=96	n=16
AVK mean (1994-2004)	83.33	83.33	83.33	83.87	81.43	87.57	86.47	88.17	87.08	90.00
±SD	9.54	9.12	9.76	9.45	9.70	9.89	8.81	11.02	10.21	11.34
AVK mean (2005-2014)	82.67	85.63	81.67	82.81	93.30	78.49	84.85	77.17	78.96	77.25
±SD	8.97	8.89	9.32	9.31	9.56	9.98	8.70	9.23	8.94	10.80
AVK mean (1994-2014)	82.93	84.30	82.09	83.19	81.39	82.38	85.83	81.23	82.18	84.24
±SD	9.46	9.09	9.59	9.40	9.67	9.95	8.71	10.08	9.81	11.02

Designations as in Table 2.

dogs (85.83 ± 8.71%) from the CH group. The lowest for male dogs from the Z group (81.39 ± 9.67%). A higher AVK means less inbreeding. The AVK coefficient increased in the group (Z) of dogs from foreign kennels to 93.30 ± 9.56% in 2005-2014. In female dogs, the highest AVK coefficient in group Z (90.00 ±11.34%) was in the analyzed period of 1994-2004.

The estimated kinship in the breed is presented by Table 6. It turned out that the average kinship between probands and their parents in the direct male line was 55.58 ± 3.42%, and in the female line 55.44 ± 3.39%. Values above 50% indicate that each line, both paternal and maternal, was characterised by a certain degree of inbreeding. A higher kinship coefficient for the paternal line may suggest that breeders look for male line inbreds.

Table 6

The proband's relationship to the parent in a straight line (mean ± SD)

Sex	Male Line	Female Line
	COR (R_{XA}) Mean±SD	COR (R_{XY}) Mean±SD
Male Dogs	55.53 ± 3.47	55.34 ± 3.49
Female Dogs	55.68 ± 3.30	55.56 ± 3.23
Total	55.58 ± 3.42	55.44 ± 3.39

COR (R_{XA}) – Primary kinship in male line;
COR (R_{XB}) – Primary kinship in female line.

Discussion

KALINOWSKA *et al.* (2010) demonstrated that half of the active population registered with the Krakow Division of ZKwP was inbred, whereas in a 4-generation population more than 23% of dogs were inbred (about 27% of males and 20% of females). The average inbreeding rate was 1.37%, and the rate estimated only among inbred animals is 5.85%. The highest inbreeding rate was 14.06%. The latest studies on the population consisting of 491 dogs showed that 46% of Tatra Shepherds (20% of males and 26% of females) were inbred, and in the standardized 4-generation population, 40%, including 16% of males and 22% of females is of an origin connected with inbreeding. The average inbreeding rate was at the level of 2.93% for the entire population and among inbred animals it was 6.34%. The highest inbreeding rate was 20.31%. The studies by KANIA-GIERDZIEWICZ & GIERDZIEWICZ (2013) conducted on the Tatra Shepherd dog breed in the Silesian voivodeship showed that 77.42% of the population is inbred (including 81.82% of males and 75% of females), and the average inbreeding rates (F_X) are respectively 4.8% for all and 5.8% for inbred animals, whereas the average kinship coefficient is 11.5%. In another experiment carried out in 2015 on the population of Tatra Shepherd dogs living in the region of Podhale, KANIA-GIERDZIEWICZ *et al.* (2015) found that the average inbreeding value for the breed is 7.17%, and the average kinship coefficient is 18.2%. In addition, the inbreeding value showed an upward trend. Similar F_X results at a level of 8.8% were obtained by LEROY *et al.* (2009) for the Pyrenean Shepherd and by CECCHI *et al.* (2016) for the Bracco Italiano breed – F_x = 6.7%. Slightly higher inbreeding levels of the Boxer breed were above 10% and the inbreeding rate amounted to 0.14% per year (MOSTERT *et al.* 2015). In contrast, Bullmastiff Dogs' inbreeding coefficients ranged from 0 in 1980 to 0.054 in 1997. An overall increase in mean inbreeding coefficient was seen until the mid 1990's, reaching 0.043 in 1995, and remained relatively stable until 0.044 in 2013 (MORTLOCK *et al.* 2016). The current Norwegian Lundehund population is highly inbred and has lost 38.8% of its genetic diversity in the base population. The ancestor with the highest contribution in the pedigree is a female with 18 offspring born in the 1960s. Her contribution to the last cohort was 41%. Immediate actions are needed to increase the genetic diversity in the current Lundehund population. The only option to secure the conservation of this rare breed is to introduce individuals from foreign breeds as breeding candidates (KETTUNEN *et al.* 2017). For Swedish protected dog breeds there is no correlation between the average *F* and the population size measured either as the size of the full pedigree or as the number of living dogs (coefficients of correlation, *r*, range from 0.00 to 0.53 and 0.15-0.60, respectively, with 0.07<P<1.00). Breeds like the Swedish Lapphund and the Swedish Vallhund, however, reach similarly high average *F* – 0.09 – in spite of the pedigree sizes of several thousand individuals, census sizes of well above 1000, and over 50 founders. Similarly, the Swedish Elkhound and the Drever have pedigrees comprising of over 50,000 dogs, with over 10,000 defined as 'alive' in 2012 but the average *F* is above 0.07 (JANSSON & LAIKRE 2018). A study of genealogical parameters for a number of breeds in Australia found that the mean inbreeding coefficient ranged from 0 to 0.101 across 32 analyzed breeds (SHARIFLOU *et al.* 2011).

The AVK of the analysed Tatra Shepherds was satisfactory and it was above 91% for the general population of dogs and 84% for the inbred population. The period of analysis significantly contributed to a reduction in AVK, mostly for female dogs from the nCH, PL, and Z groups born in 2005-2014. Similar results for AVK were obtained for Newfoundland dogs, i.e. above 85% (KRUZIŃSKA *et al.* 2019).

Secondary kinship R_{XY} within the entire population was lower than for inbred pairs by 4.25%. On the other hand, similar results were obtained for pairs with 4-generation pedigrees. KALINOWSKA *et al.* (2010) obtained similar R_{XY} values for inbred pairs

within the range 14.51-14.92% and lower for the entire population, 4.53-6.12%. Slightly lower R_{XY} results, around 3.91%, were estimated for the entire population of German Shepherd dogs from the region of Krakow (KANIA-GIERDZIEWICZ *et al.* 2011).

With regard to quite a high level of inbreeding in the breed, the introduction of new blood is recommended in breeding Tatra Shepherd dogs. The sources used to enrich the gene pool can be dogs entered in the stud book. Insofar as excessive inbreeding can contribute to inbreeding depression, the dogs entered in the stud book, despite their phenotype complying with the breed standard, are a mystery in terms of origin, genotype, possible inbreeding or why their ancestors were excluded from breeding. Thus, dogs to be entered into the stud book should be thoroughly selected and these should only be dogs having unquestionable typical physical and mental traits of the breed. Among all the stud dogs in Poland mentioned in the Newsletter of the Tatra Shepherd Dogs Club, 61.11% did not leave a male line continuer (POLISH KENNEL CLUB 2009; KALINOWSKA *et al.* 2010). A way of enriching the line can be reaching the descendants of dogs that are not shown at dog shows and obtaining genetic material by entering them into the stud book and then using them for breeding. Therefore, the import of Tatra Shepherd dogs bred in the Netherlands or France that show a low degree of kinship with dogs in Poland can be an excellent choice to abandon extensive inbreeding, and aim at improving the breed (SELL 2009; JANNSON & LAIKRE 2014; KANIA-GIERDZIEWICZ *et al*. 2015; RADKO *et al.* 2018).

Conclusion

The mean F_x inbreeding coefficient in the analyzed period was 2.93%, including 3.14% of males and 2.79% of females. The greatest inbreeding was observed in males who did not have the title of champion (nCH) and females born in Poland (PL).

The average AVK level of ancestor loss value in the analyzed period in the group of males was 82.93%, and in the group of females – 82.38%. The lowest AVK index was found in the group of dogs bred abroad, and the highest in the group of champion dogs.

Taking into account the inbreeding rate of the breed, it was concluded that there is not yet a risk of inbreeding depression, but there is a downward trend in the diversity of the gene pool. Therefore, it is suggested that the stud book for the Tatra Shepherd dog is left open because the breed has gained popularity year on year, so breeding will increase.

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Author Contribution

A Research concept and design: E.S., E.H., R.N.; B Collection and/or assembly of data: E.S.; Data analysis and interpretation: E.S., E.H., R.N.; Writing the article: E.S., E.H., R.N.; Critical revision of the article: E.H., R.N.; Final approval of article: E.S., E.H., R.N.

Conflict of Interest

The authors declare that there is no conflict of interest between them and other people or with organizations that could inappropriately bias the results.

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