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Transferrin genotypes as genetic markers of lifetime prolificacy of ewes in a flock of prolific sheep 09

Genotypy transferyny jako genetyczne markery całożyciowej plenności maciorek w stadzie owcy plennej 09

Summary. The aim of the study was to investigate the possibility to use polymorphic variants of transferrin (Tf) as genetic markers of prolificacy in ewes in the first six breeding seasons. The experimental material consisted of 89 ewes of prolific sheep 09 (44% Polish Merino, 31% East-Friesian milk sheep, 25% Finnish sheep). The mean size of the litter from ewes having different transferrin genotypes in individual periods within the 6-year-long reproduction utilization exhibited considerable variability. In the whole investigated period, covering six breeding seasons, the most prolific ewes were those with transferrin genotypes BB and DP, with the mean litter size of 1.70. In all the five analyzed periods there is a clear trend for ewes with transferrin genotype MM to have the lowest index values. When selecting the flock of prolific sheep line 09 for increased prolificacy the transferrin genotype may be applied as an additional selection criterion and it would be advisable to keep for the purpose of flock replacement ewes with genotypes Tf BB or Tf DP, while ewes with genotype Tf MM, as the least prolific, should be culled.

Key words: sheep, prolificacy, genetic markers, transferrin

INTRODUCTION

Prolificacy of ewes affects the number of produced lambs and determines the efficiency of mutton production in sheep farming. At a low level of this trait in the Polish sheep population, an important breeding objective is to achieve genetic improvement of prolificacy through selection. Since direct selection on the basis of litter size is not very effective, other selection criteria are searched for, e.g. genetic markers of this trait.

Transferrin, one of class I genetic markers, is the most heterogeneous polymorphic blood protein in sheep [Kurył 1992]; a total of 12 codominant alleles have been found in its locus. This protein, belonging to the group of beta-globulins, is found not only in blood serum, but also milk and semen. The main function of transferrin in the organism is to participate in iron metabolism and in immune responses. A review of the functions of this protein in the organism is given by Kmieć [1998]. Considerable polymorphism and various biological functions of transferrin resulted in a situation when it is used e.g. in studies on the genetic structure of the population, to estimate genetic distance, whereas genotypes of this protein may be used as genetic markers of production traits and markers of immunity/susceptibility to diseases in sheep.

The aim of this study was to investigate the possibility of using polymorphic variants of transferrin as genetic markers of prolificacy in ewes in the first six breeding seasons. Finding a link between transferrin genotypes in a flock and prolificacy of ewes may facilitate selection for further breeding of these ewes and rams, which would exhibit high prolificacy.

MATERIAL AND METHODS

The experimental material consisted of 89 ewes of prolific sheep 09 (44% Polish Merino, 31% East-Friesian milk sheep, 25% Finnish sheep) formed and kept on the Złotniki farm, belonging to the Agricultural University of Poznań. Tested ewes (n = 89) were born in the years 1990–1996. The type of birth of these ewes and the prolificacy of their dams were adopted as selection criteria when selecting animals for flock replacement. Replacement ewes came from twin or triple births; at the same time it was attempted to ensure that lifetime prolificacy of their dams exceeded the flock mean. Ewes were serviced for the first time in the first year of life and in successive seasons they were used in breeding once a year.

Transferrin (Tf) genotypes were determined in two stages. In the first the method of starch gel horizontal electrophoresis was applied according to Smithies [1995] as modified by Bojczuk *et al.* [1980]. In the second stage in order to precisely distinguish allele Tf^{D} from allele Tf^{M}, genotypes were determined in zonal gradient of polyacrylamide gel [Gahne *et al.* 1977].

Allele frequencies and percentages of individual transferrin genotypes were calculated for the flock. High polymorphism of transferrin was manifested in the occurrence of six alleles in the flock and determined the occurrence of 15 genotypes of this protein. For the purpose of the analysis of variance the genotypes, the number of which within individual traits was lower than 5, were excluded from further analysis.

The following breeding performance traits were investigated:

– the number of lambs born in the 1st, 2nd, 3rd, 4th, 5th and 6th breeding seasons; for barren sheep in individual seasons the assumed number of lambs was 0,

– the total number of lambs born in the following periods of breeding performance: seasons 1 + 2 (period 1–2), 1 + 2 + 3 (period 1–3), 1 + 2 + 3 + 4 (period 1–4), 1 + 2 + 3 + 4 + + 5 (period 1–5), 1 + 2 + 3 + 4 + 5 + 6 (period 1–6),

– the mean litter size, expressed as the ratio of the number of born lambs to the number of lambings (excluding the years in which the sheep was barren), in the above mentioned periods.

Values of the investigated breeding traits are expressed in a discrete scale. Thus, probit transformation of discrete values was applied and a multivariate analysis of variance was performed on the transformed data using the least squares method [SAS 1989]. Means given in tables are arithmetic means.

In order to determine the effect of transferrin genotype, as well as other fixed and random factors on the analyzed reproduction traits, the following linear model was applied:

$$y_{ijklm} = \mu + a_i + b_j + c_k + s_l + e_{ijklm}$$

where:

y_{ijklm} – phenotypic value of a trait,
μ – total mean,
a_i – fixed effect of i-th transferrin genotype (i = 1, 2...8),
b_j – fixed effect of j-th birth type of the dam (j = 1, 2, 3),
c_k – fixed effect of k-th year of birth of the dam (k = 1, 2...7),
s_l – random effect of l-th sire (l = 1, 2...16),
e_{ijklm} – effect of random error connected with ijklm-th observation.

RESULTS AND DISCUSSION

In the analyzed flock a total of 6 transferrin alleles were identified (tab. 1). Alleles with the highest frequencies were Tf^D (q = 0.3146) and Tf^B (q = 0.2416). The lowest frequency was found for allele Tf^P (q = 0.0449). The above mentioned 6 alleles determined the occurrence of 15 transferrin genotypes, characterized by varying frequencies (tab. 1). Four genotypes, i.e. Tf AD, Tf BC, Tf BD and Tf CD, were genotypes found in the biggest number of the tested ewes – a total of 44 ewes. The least frequent genotypes were Tf AM (n = 1), as well as Tf AC, Tf BP and Tf CC (for each genotype n = 3).

Table. 1 Frequency of transferrin alleles and genotypes in the investigated flock
Tabela 1. Częstość występowania alleli i genotypów transferyny w badanym stadzie

Alleles Allele	n	q	Genotypes Genotypy	n	%
Tf^A	18	0.1011	AB	4	4.5
Tf^B	43	0.2416	AC	3	3.4
Tf^C	32	0.1798	AD	10	11.2
Tf^D	56	0.3146	AM	1	1.1
Tf^M	21	0.1180	BB	5	5.6
Tf^P	8	0.0449	BD	11	12.4
			BC	11	12.4
			BM	4	4.5
			BP	3	3.4
			CC	3	3.4
			CD	12	13.5
			DD	7	7.9
			DM	4	4.5
			DP	5	5.6
			MM	6	6.6
Sum Suma	178	1.0000	Sum Suma	89	100.00

Table 2 presents results showing the mean number of lambs born by dams with individual transferrin genotypes in each season, as well as the effect of the type of birth and the year of birth of ewes on the level of the investigated traits. In the first breeding season the biggest number of lambs were born by ewes with transferrin genotype AD – 1.80, while the lowest number by ewes with genotype Tf BD – 1.18 ($P \leq 0.05$). In the second season the biggest number of lambs were born by ewes with genotypes Tf BB, Tf BD and Tf BC, in which the mean number of born lambs was by at least 1.05 ($P \leq 0.01$) higher than the figure for ewes with transferrin genotype MM, in which the lowest level of this index was found for this trait (0.85). Also, in the 4th and 5th breeding seasons ewes with genotype Tf MM had the lowest mean number of born lambs, i.e. 1.71 and 1.42, respectively. In seasons 3, 4 and 5, the highest indexes were observed for ewes with different transferrin genotypes. In the 6th season the highest value of this index was found for ewes with genotype Tf BB, although this result was not confirmed statistically.

The type of birth of ewes had a significant effect only on the number of lambs born in the 1st breeding season ($P \leq 0.05$). A significant effect of the year of birth of ewes was found on the number of born lambs in each, except for the third breeding season.

Table 3 gives means presenting the total number of lambs born by the tested ewes in five periods within six breeding seasons. In the first four breeding seasons combined (period 1–4), the biggest number of lambs were born by ewes with genotypes Tf BC, Tf BD and Tf CD (7.30, 7.36 and 7.41, respectively). The lowest value of this index was obtained for ewes with genotype Tf MM. An identical trend was found for period 1–5. The difference between means for ewes with genotypes Tf MM and means for ewes with the other transferrin genotypes were confirmed statistically, both for periods 1–4 and 1–5. The total number of lambs born in the whole 6-year-long period (1–6) was the highest in ewes with genotype Tf BD (on average 11.27), but ewes with genotypes Tf BB and Tf BC also reached high index values (11.00 for each genotype). In the same period ewes with genotype Tf MM obtained the lowest level of this index – a mean 8.75 born lambs. No effect of the type of birth of ewes was found on the number of lambs they had in the analyzed periods of their lives.

The mean litter size for ewes in individual periods varied considerably (Tab. 4). Ewes with genotypes Tf BD and Tf CD reached the highest index values in period 1–4, but in period 1–5 ewes with genotypes Tf AD and Tf DP were most prolific. In the whole analyzed period, covering six breeding seasons, ewes with transferrin genotypes BB and DP were most prolific, as their mean litter size was 1.70. In all the five analyzed periods a trend may be observed for ewes with transferrin genotype MM to reach the lowest index values. Differences between means for ewes with genotype Tf MM and means for ewes with other transferrin genotypes were confirmed statistically first of all in period 1–5 ($P \leq 0.01$, $P \leq 0.05$). In periods 1–2 and 1–4 differences between means for ewes with genotypes TF BD and Tf MM were confirmed statistically (1.90 vs. 1.28 in period 1–2 and 1.96 vs. 1.57 in period 1–4). No effect of the type of birth of ewes was found on litter size in any of the analyzed periods. A significant effect of the year of birth of ewes ($P \leq 0.05$) was observed in periods 1–4, 1–5 and 1–6.

One of the first publications in the Polish animal science literature concerning the effect of transferrin and hemoglobin genotypes on prolificacy of ewes was a study by Bojczuk and Bojczuk [1988]. Those authors, while investigating two flocks of Polish lowland sheep, did not find a dependency between genotypes of the analyzed markers and twin litters obtained from ewes. Kmieć [1991], while studying Polish long-wooled ewes, found that in the first four breeding seasons ewes with transferrin genotype DD exhibited the highest prolificacy, whereas ewes with genotypes Tf AD and Tf DE were animals with the highest fertility.

Table. 2 The effect of transferrin genotype, type and year of birth of dams on the number of lambs born in each season
Tabela 2. Wpływ genotypu transferyny, typu i roku urodzenia matek na liczbę jagniąt urodzonych w poszczególnych sezonach

Factor Cynnik	n	Season 1 Sezon 1 $\bar{x}$ ± SD	Season 2 Sezon 2 $\bar{x}$ ± SD	Season 3 Sezon 3 $\bar{x}$ ± SD	Season 4 Sezon 4 $\bar{x}$ ± SD	Season 5 Sezon 5 $\bar{x}$ ± SD	Season 6 Sezon 6 $\bar{x}$ ± SD
Genotype of Tf							
Genotyp Tf							1.40 ± 0.84
AD	10	1.80 ± 0.42 a	1.50 ±0.52 Aa	1.90 ± 0.56	1.80 ± 0.63	1.90 ± 0.73 A	2.00 ± 0.0
BB	5	1.40 ± 0.54	2.00 ± 0.0 Ba	2.00 ± 0.0	1.80 ± 0.44	1.80 ± 0.44 a	1.80 ± 0.91
BC	10	1.40 ± 0.51	1.90 ± 0.31 C	2.00 ± 0.47	2.00 ±0.66	1.90 ± 0.31 b	1.81 ± 0.40
BD	11	1.18 ± 0.75 a	2.09 ± 0.53 AD	1.81 ± 0.40	2.27 ± 0.78	2.09 ± 0.30 Babc	1.41± 0.66
CD	12	1.50 ± 0.67	1.75 ± 0.62 EF	2.00 ± 0.42	2.16 ± 0.71	1.91 ± 0.79 cd	1.71 ± 1.11
DD	7	1.42 ± 0.53	1.57 ± 0.53 G	1.85 ± 0.37	2.14 ± 0.37 a	2.13 ± 0.36 e	1.80 ± 0.83
DP	5	1.60 ± 0.54	1.80 ± 0.44 a	2.00 ± 0.0	1.60 ± 0.54	2.00 ± 0.0	1.64 ± 0.69
MM	7	1.28 ± 0.48	0.85±0.89 BCDEFGa	1.85 ± 0.37	1.71 ± 0.75 a	1.42 ± 0.78 ABde	
Type of birth Typ urodzenia		*	ns	ns	ns	ns	ns
Year of birth Rok urodzenia		*	*	ns	**	*	*

Within the columns, within factors, values designated with the same capital (small) letters differ significantly at $P \leq 0.01$ ($P \leq 0.05$); ** $P \leq 0.01$, * ($P \leq 0.05$), ns – insignificant
W kolumnach, w ramach czynników, wartości oznaczone tą samą dużą literą (małą) różnią się istotnie przy $P \leq 0{,}01$ ($P \leq 0{,}05$); ** $P \leq 0{,}01$, * ($P \leq 0{,}05$); ns – nieistotne

Table 3. The effect of transferrin genotype, type and year of birth of dams on the total number of lambs born in each period
Tabela 3. Wpływ genotypu transferyny, typu i roku urodzenia matek na łączną liczbę jagniąt urodzonych w poszczególnych okresach

Factor Czynnik	n	Period 1–2 Okres 1–2 $\bar{x} \pm SD$	Period 1–3 Okres 1–3 $\bar{x} \pm SD$	Period 1–4 Okres 1–4 $\bar{x} \pm SD$	Period 1–5 Okres 1–5 $\bar{x} \pm SD$	Period 1–6 Okres 1–6 $\bar{x} \pm SD$
Genotype of Tf Genotyp Tf						
AD	10	3.30 ± 0.48 A	5.20 ± 0.63 a	7.00 ± 0.94 a	8.90 ± 0.99 A	10.30 ± 1.56 Aa
BB	5	3.40 ± 0.54 B	5.40 ± 0.54 A	7.20 ± 0.83 A	9.00 ± 0.70 B	11.00 ± 0.70 b
BC	10	3.30 ± 0.67 C	5.30 ± 0.82 B	7.30 ± 0.94 B	9.20 ± 1.13 C	11.00 ± 1.56 a
BD	11	3.27 ± 0.46 D	5.09 ± 0.70 C	7.36 ± 1.02 C	9.45 ± 1.03 D	11.27 ± 1.00 AB
CD	12	3.25 ± 0.96 E	5.25 ± 1.13 D	7.41 ± 1.16 D	9.33 ± 1.72 E	10.75 ± 1.65 c
DD	7	3.00 ± 0.81 F	4.85 ± 0.89 b	7.00 ± 1.00 E	9.14 ± 1.21 F	10.85 ± 1.67 d
DP	5	3.40 ± 0.89 a	5.40 ± 0.89 c	7.00 ± 0.70 b	9.00 ± 0.70 G	10.80 ± 0.44 e
MM	7	2.14 ± 1.06 ABCDEFa	4.00 ± 1.00 ABCDabc	5.71 ± 1.60 ABCDEab	7.14 ± 1.21 ABCDEFG	8.75 ± 1.38 Bbcde
Type of birth Typ urodzenia		ns	ns	ns	ns	ns
Year of birth Rok urodzenia		*	ns	*	**	*

Within the columns, within factors, values designated with the same capital (small) letters differ significantly at $P \leq 0.01$ ($P \leq 0.05$); ** $P \leq 0.01$, * ($P \leq 0.05$), ns – insignificant
W kolumnach, w ramach czynników, wartości oznaczone tą samą dużą literą (małą) różnią się istotnie przy $P \leq 0,01$ ($P \leq 0,05$); ** $P \leq 0,01$, * ($P \leq 0,05$); ns – nieistotne

Later studies by that author [1997] confirmed the possibility to indicate in the analyzed flock of sheep these ewes, in which prolificacy depending on their transferrin genotype was significantly higher. Previous studies by Steppa and Ślósarz [1999] on sheep from the same flock (prolific sheep line 09) showed that jointly in the first three breeding seasons ewes with transferrin allele Tf^B or Tf^M ovulated the highest number of egg cells (6.25 and 6.38). In ewes with allele Tf^B the lowest embryonic losses were found (0.87), determined on the basis of the difference between litter size and the number of ovulated egg cells. These losses in ewes having allele Tf^M amounted to the average of 1.36 egg cells. Darcan and Gűney [2001], in Awassi and Cukurova Assaf sheep (5/8 Awassi, 3/8 East-Friesian sheep) kept under subtropical conditions did not find the effect of transferrin alleles on litter size. Their studies were conducted on a small flock of 40 ewes, which could have affected the obtained results.

Table 4. The effect of transferrin genotype, type and year of birth of dams on the mean litter size in each period

Tabela 4. Wpływ genotypu transferyny, typu i roku urodzenia matek na średnią wielkość miotu w poszczególnych okresach

Factor Czynnik	n	Period 1–2 Okres 1–2 $\bar{x} \pm SD$	Period 1–3 Okres 1–3 $\bar{x} \pm SD$	Period 1–4 Okres 1–4 $\bar{x} \pm SD$	Period 1–5 Okres 1–5 $\bar{x} \pm SD$	Period 1–6 Okres 1–6 $\bar{x} \pm SD$
Genotype of Tf Genotyp Tf						
AD	10	1.65 ± 0.24	1.73 ± 0.21	1.75 ± 0.23	1.85 ± 0.41^{A}	1.60 ± 0.51
BB	5	1.70 ± 0.27	1.80 ± 0.18	1.80 ± 0.20	1.60 ± 0.22	1.70 ± 0.27^{ab}
BC	10	1.65 ± 0.33	1.76 ± 0.27	1.82 ± 0.23	1.65 ± 0.33	1.60 ± 0.51
BD	11	1.90 ± 0.59^{a}	1.84 ± 0.38	1.96 ± 0.42^{a}	1.63 ± 0.39^{B}	1.50 ± 0.31
CD	12	1.70 ± 0.45	1.80 ± 0.36	1.89 ± 0.27	1.70 ± 0.65^{a}	1.45 ± 0.49^{a}
DD	7	1.50 ± 0.40	1.61 ± 0.30	1.75 ± 0.25	1.78 ± 0.39^{b}	1.57 ± 0.67
DP	5	1.70 ± 0.44	1.80 ± 0.29	1.75 ± 0.17	1.80 ± 0.27	1.70 ± 0.44
MM	7	1.28 ± 0.26^{a}	1.54 ± 0.12	1.57 ± 0.22^{a}	1.35 ± 0.37^{ABab}	1.41 ± 0.56^{b}
Type of birth Typ urodzenia		ns	ns	ns	ns	ns
Year of birth Rok urodzenia		ns	ns	*	*	*

Within the columns, within factors, values designated with the same capital (small) letters differ significantly at $P \leq 0.01$ ($P \leq 0.05$); ** $P \leq 0.01$, * ($P \leq 0.05$), ns – insignificant

W kolumnach, w ramach czynników, wartości oznaczone tą samą dużą literą (małą) różnią się istotnie przy $P \leq 0{,}01$ ($P \leq 0{,}05$); ** $P \leq 0{,}01$, * ($P \leq 0{,}05$), ns – nieistotne

The high mean litter size in ewes with transferrin genotype BB shown in this study confirms the results of previous studies [Steppa 2005], in which prolificacy of 249 ewes of this flock was analyzed in the first four breeding seasons.

CONCLUSIONS

1. Selecting a flock of prolific sheep line 09 for improved prolificacy the transferrin genotype may be applied as an additional selection criterion and those ewes which have genotypes Tf BB or Tf DP should be left for the purpose of flock replacement.
2. Ewes with genotype Tf MM, as least prolific, should be culled.

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Streszczenie. Celem pracy było sprawdzenie możliwości wykorzystania polimorficznych wariantów transferyny (Tf) jako genetycznych markerów plenności maciorek w pierwszych sześciu sezonach rozpłodowych. Materiałem badawczym było 89 maciorek owcy plennej 09 (44% merynos polski, 31% wschodniofryzyjska owca mleczna, 25% owca fińska). Średnia wielkość miotu maciorek mających różne genotypy transferyny w poszczególnych okresach, w ramach sześciu lat użytkowania rozpłodowego, wykazywała dużą zmienność. W całym badanym okresie, obejmującym sześć sezonów rozpłodowych, za najplenniejsze można uznać maciorki mające genotypy transferyny BB i DP, które uzyskały średnią wielkość miotu 1,70. We wszystkich pięciu analizowanych okresach widoczna jest tendencja do osiągania najniższych wielkości wskaźników przez maciorki z genotypem transferyny MM. Selekcjonując stado owiec plennych 09 w kierunku podniesienia plenności, można jako dodatkowe kryterium selekcji zastosować genotyp transferyny i pozostawiać do remontu stada maciorki mające genotypy Tf BB lub Tf DP, jednocześnie maciorki mające genotyp Tf MM, jako najmniej plenne, powinny być brakowane.

Słowa kluczowe: owce, plenność, markery genetyczne, transferyna