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Description of the *Anser cygnoides* Goose Karyotype

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The karyotype of the domestic goose *A. cygnoides* was characterised on the basis of R and C bands. Chromosomal preparations obtained from an *in vitro* culture of blood lymphocytes were stained by means of the RBG and CBG banding techniques. The first nine pairs of chromosomes were analysed by the R banding technique, while fourteen pairs of chromosomes were analysed by the C banding technique. The localisation of R bands as well as the sizes and positions of constitutive heterochromatin blocks were determined. Ideograms of R and C banded patterns for the analysed chromosomes were drawn. The morphological make-up of the analysed chromosomes was assessed.

Key words: *Anser cygnoides* goose, karyotype, R bands, C bands

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Two species of the *Anser* genus comprise the ancestors of the domestic goose: the European Grey-leg goose *A. anser* and the Asian Swan goose *A. cygnoides* (CRAWFORD 1990). Crosses of both species give fertile progeny (MAZANOWSKI *et al.* 1985). The karyotypes of birds originating from *A. anser* and *A. cygnoides* show a morphological difference in the fourth pair of macrochromosomes (BHATNAGAR 1968; SHOFFNER *et al.* 1979; APITZ *et al.* 1995; HIDAS 1993, 1999) and further generations of crosses are characterised by a polymorphic stage of the fourth pair of chromosomes (RABSZTYN *et al.* 1998). Despite these investigations, a standard karyotype for *A. cygnoides* is not available. Only a standard band pattern for the domestic chicken limited to the first nine pairs of chromosomes has been published (LADJALI-MOHAMMEDI *et al.* 1999).

This work aimed at describing the karyotype of *A. cygnoides* by RBG and CBG techniques of chromosome staining.

Material and Methods

Peripheral blood of domestic geese originating from *A. cygnoides* sampled from ten birds was analysed. Two traditional banding techniques: RBG (PERRY & WOLFF 1974) and CBG (SUMNER 1972) were applied. An *in vitro* lymphocyte culture was carried out in the case of the RBG technique. It included the incorporation of BrdU and Hoechst 33258 in the 65th hour of incubation, as well as EB and colchicine in the 69th hour of incubation. Ten metaphase plates were analysed for each bird. Analysis of chromosomes stained with the RBG technique made it possible to draw a repeating band pattern of nine chromosomes presented in the form of ideograms, including the layout of characteristic light and dark bands. The description of chromosomal band patterns was prepared according to widely accepted principles (ISCNDA 1989, LADJALI-MOHAMMEDI *et al.* 1999).

The application of the CBG technique led to the determination of the C band pattern on chromosomes. Moreover, the size of individual heterochromatin bands was measured and expressed as a relative value in relation to the whole length of the chromosome. Constitutive heterochromatin blocks located on the p and q arms of heterochromosome W were measured as one large block because the light band situated in the centromeric part of the chromosome was very thin. The results obtained were statistically described. The index of arms [q/p] and centromeric index [p/(p+q)] were calculated for the following chromosomes: the first, second, third autosome and sex chromosomes Z W. Acromeric chromosomes in which only the q arm was visible were not measured.

Results

Somatic cells of the goose *A. cygnoides* include eighty chromosomes of different size (Fig. 1). The largest chromosomes were from 4 to 15 μm long, depending on the metaphase plate, whereas the length of the smallest ones ranged from 0.68 to 1.80 μm.

A band pattern ideogram of eight pairs of autosomes and sex chromosomes stained with the RBG technique (Fig. 2) was prepared. Altogether a hundred and ten R bands were found, including fifty-eight positive dark bands and fifty-two negative light ones.

The chromosome of the first pair was classified as submetacentric and had an index of arms equal to 1:1.704 and centromeric index of 0.377. Twenty-nine R bands were detected. Two regions with eleven R bands, including six positive ones, were determined on the p arm. On the q arm of the chromosome, eighteen R bands, nine positive and nine negative, were found within three regions.

The second submetacentric chromosome had an arms index of 1:1.505 and centromeric index equal to 0.405. Twenty-six R bands were determined. Two regions were found on the p arm. They included twelve R bands, half of which were positive. The q arm of the chromosome contained two regions with fourteen R bands, including seven positive ones.

The index of arms of the third acrocentric chromosome was 1:5.017, whereas the centromeric index was 0.171. Twenty R bands were found. Three R bands were observed within one region on the p arm of the chromosome. An interstitial dark band was located between two light negative ands. On the q arm of the chromosome, two regions with seventeen R bands, including nine positive and eight negative ones, were determined.

The value of the centromeric index calculated for the fourth metacentric autosome was 0.459, whereas the index of arms was 1:1.110. Nine R bands were found. Two dark bands separated by a light band were observed on the p arm. The q arm of the aforementioned chromosome had one region with six R bands, half of which were positive.

The acrocentric chromosome of the fifth pair contained ten R bands. One region with one positive band was found on the p arm, whereas on the q arm there was one region which included nine bands, five of which were positive while the remaining four bands were negative.

Seven R bands were observed within the q arm of the sixth acrocentric chromosome, including four dark positive bands.

The seventh acrocentric autosome had nine R bands out of which five were positive and four were negative.

Eight R bands were found on the eighth acrocentric autosome, including five dark positive bands and four light negative ones.

Submetacentric sex chromosome Z had an index of arms 1:1.351 and centromeric index equalling 0.399. Seventeen R bands were recorded. One region was found on the p arm and it included two

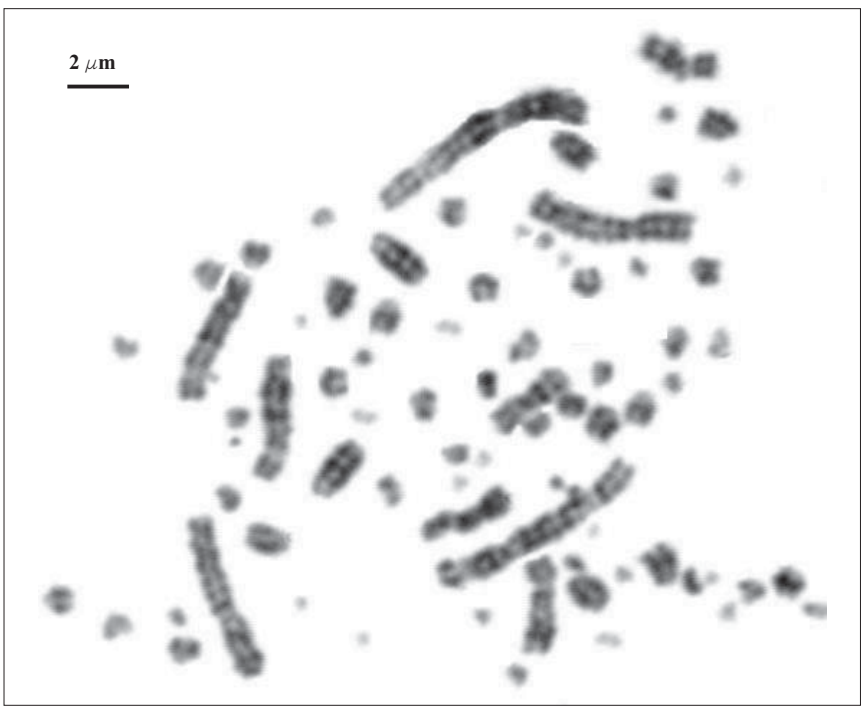

Fig. 1. Metaphase plate of the chromosomes of *Anser cygnoides* (RBG banding).

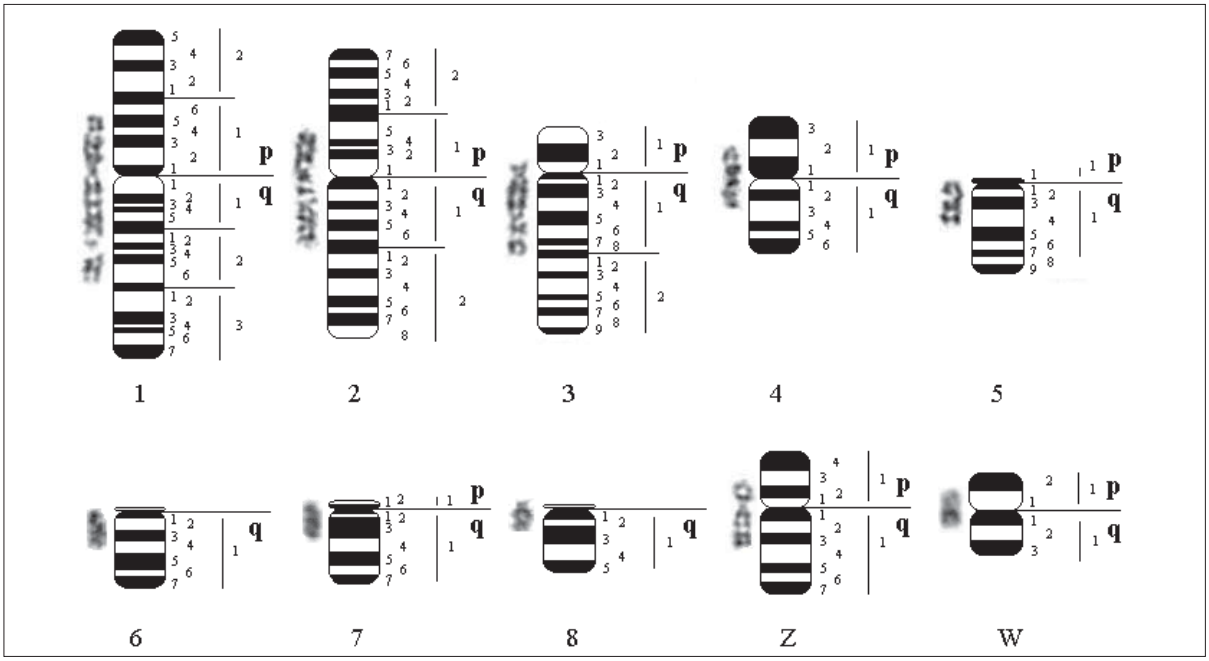

Fig. 2. Ideogram and karyogram of the chromosomes of *Anser cygnoides* (RBG banding).

dark bands and two light bands. On the q arm one region was observed containing four positive and three negative bands.

Sex chromosome W had an index of arms 1:1.395 and centromeric index 0.405. Five R bands were recorded. One region was determined on the p arm. In the distal part of the chromosome a positive band was found, whereas in the proximal part the recorded band was negative. One region of the q arm yielded positive bands in its proximal part, negative interstitial bands and positive bands in its distal part.

Among the forty pairs of chromosomes, constitutive heterochromatin was identified on the thirteen largest chromosomes and on the pair of sex chromosomes. (Fig. 3). Twenty-five heterochromatin blocks were observed on the analysed chromosomes (Fig. 4). Constitutive heterochromatic regions in the proximal parts of p and q arms were observed on all the examined chromosomes, ex-

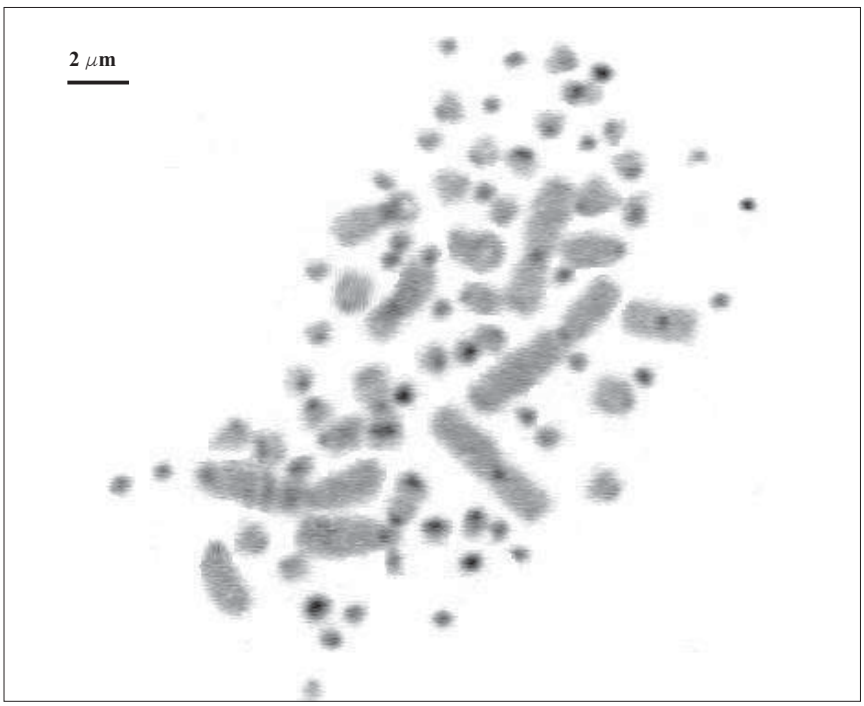

Fig. 3. Metaphase plate of the chromosomes of *Anser cygnoides* (CBG banding).

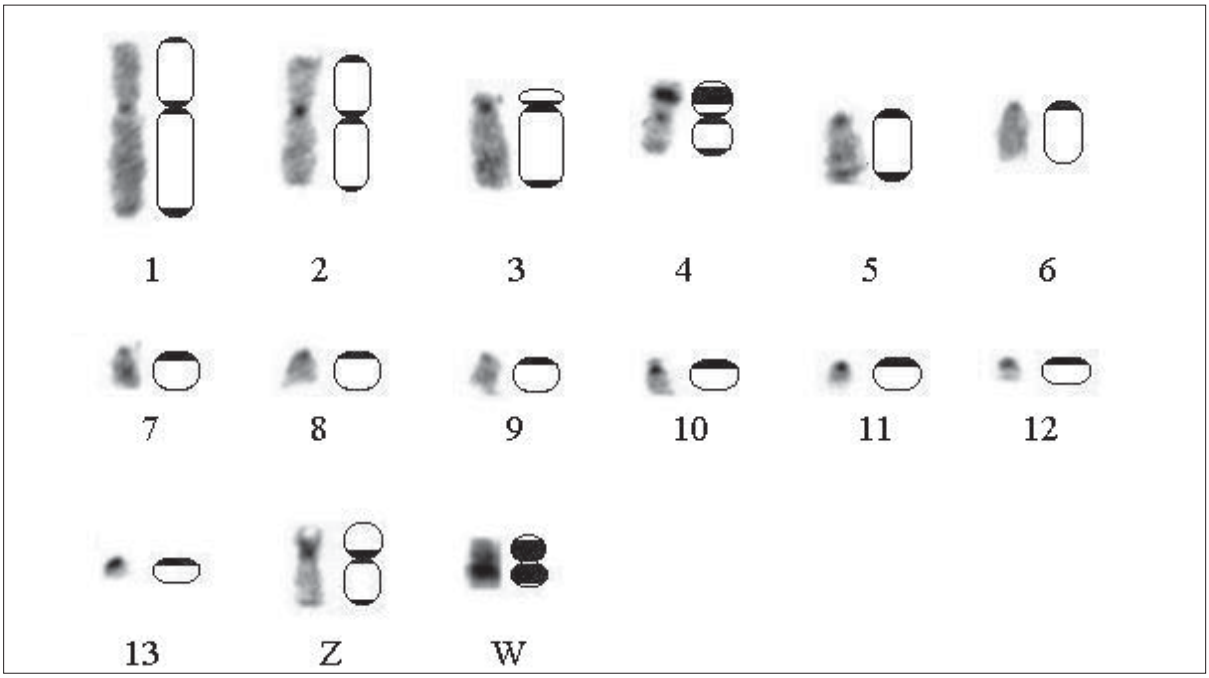

Fig. 4. Ideogram of the chromosomes of *Anser cygnoides* (CBG banding).

Table 1

Size of constitutive heterochromatin blocks of thirteen autosomes and sex chromosomes of *Anser cygnoides*

Chromosome	Chromosome region – Statistical description				
	Proximal	Distal		Interstitial	
	$\overline{X} \pm S$	p arm	q arm	p arm	q arm
		$\overline{X} \pm S$		$\overline{X} \pm S$	
1	8.140±0.287	5.481±0.266	7.399±0.325	–	–
2	9.216±0.723	5.390±0.370	8.155±0.530	–	–
3	13.314±0.569	–	12.424±0.491	–	–
4	9.503±0.860	–	15.249±1.333	18.394±0.697	–
5	14.409±1.047	–	16.411±1.072	–	–
6	18.377±4.528	–	–	–	–
7	22.659±3.440	–	–	–	–
8	24.662±4.147	–	–	–	–
9	25.200±4.318	–	–	–	–
10	27.030±6.063	–	–	–	–
11	28.777±6.459	–	–	–	–
12	30.565±4.662	–	–	–	–
13	31.605±5.433	–	–	–	–
Z	13.579±0.784	–	6.475±0.869	–	–
W	–	–	–	74.830±4.588	

cluding chromosome W. The remaining constitutive heterochromatin blocks were found in the distal parts of both arms of the first and second chromosome pairs, and on the q arms of the third, fourth and fifth autosomes. In contrast, a telomeric band on chromosome Z was observed only on the long arm. In addition, a heterochromatin block was found in the interstitial part of the q arm of the fourth chromosome and in the same part of the p and q arms of chromosome W.

Average values of the sizes of the heterochromatin blocks ranged from 18 to 32% (Table 1). The smallest amount (18%) of constitutive heterochromatin, compared with other analysed chromosomes, was

detected on the sixth autosome, whereas the largest amount (75%) was found on sex chromosome W.

Discussion

The morphological make-up of the chromosomes of the goose *A. cygnoides* was described in several studies. The characterisation of the morphology of *A. cygnoides* presented by SILVERSIDES *et al*. (1988), HIDAS (1993, 1999), and APITZ *et al.* (1995) is in accordance with the results of the present work. Chromosomes of the first and second pair as well as sex chromosomes were classified as submetacentric, four chromosome pairs as metacentric and the remaining autosomes as acrocentric. The values of centromeric indices presented by Apitz and co-workers are similar to the results obtained in this study.

There is no data on the RBG banding pattern of *A. cygnoides* chromosomes. SILVERSIDES *et al.* (1988) and APITZ *et al.* (1995) analysed band patterns of the first five autosomes and sex chromosomes of *A. cygnoides* stained by the GTG technique. APITZ *et al.* (1995) observed thirty-two dark positive G bands on the examined chromosomes. LADJALI *et al.* (1995) analysed hen chromosomes stained by the method of R and G bands and found that R bands do not reflect the commonly accepted principle of reversed G bands. The band pattern standard for *Gallus domesticus* prepared by LADJALI-MOHAMMEDI *et al.* (1999) is a reference point for studies on other bird species (SCHMID *et al.* 2000). In the present study seventy R-positive bands were found, which is the same as the amount for *Gallus gallus* (LADJALI-MOHAMMEDI *et al.* 1999).

The constitutive heterochromatin blocks detected in the proximal and interstitial chromosomal parts in *A. cygnoides* presented in the form of an ideogram (Fig. 4) are in accordance with those presented by APITZ *et al.* (1995). The authors of the aforementioned studies do not mention the localisation of heterochromatin in the distal parts of chromosomes. HAMMAR *et al.* (1966) observed C bands in the telomeric regions. Heterochromatin blocks in the distal parts of chromosomes do not stain as well as the pericentromeric bands. The detection of C bands in this part of a chromosome requires a longer duration of chromatin digestion by barium hydroxide.

The measurements of heterochromatin block sizes carried out in this study and expressed as relative values in relation to the chromosomal length made it possible to determine the variability of chromosomal construction. The size of constitutive heterochromatin blocks in different animal species were evaluated by SŁOTA (1988), ŚWITOŃSKI (1988) and KOZUBSKA-SOBOCIŃSKA *et al.* (1999).

References

APITZ M., WAGNER K. U., SAAR W. 1995. Karyotype characteristics in domestic ducks and geese. Proc. 10th Europ. Symp. Waterfowl, Halle, Germany: 465-472.

BHATNAGAR M. K. 1968. Mitotic chromosomes of White Chinese Geese. J. Hered. **59**: 191-195.

CRAWFORD R. D. 1990. Origin and history of poultry species. (In: Poultry Breeding and Genetics, ed. Elsevier, Amsterdam-Oxford-New York-Tokyo): 1-41.

HAMMAR B. 1966. The karyotypes of nine birds. Hereditas **55**: 367-585.

HIDAS A. 1993. Cytogenetic studies on a species hybrid goose breed. Proc. 8th North American Colloquium on Domestic Animal Cytogenetic and Gene Mapping, Guelph, Canada: 153-155.

HIDAS A. 1999. Molecular cytogenetic studies in domestic goose. Proc. 13th European Colloquium on Cytogenetics of Domestic Animals, Budapest, Hungary. Allatenyesztes es Takarmanyozisenetics **48**: 78-80.

ISCNDA 1989. International System for Cytogenetic Nomenclature of Domestic Animals (1989). The Second Internat. Conf. Standardization of Domestic Animal Karyotypes. Di Berardino D., Hayes H., Fries R., Long S. 1990. Cytogenet. Cell Genet. **53**: 65-79.

KOZUBSKA-SOBOCIŃSKA A., SŁOTA E., BUGNO M., DANIELAK-CZECH B., REJDUCH B. 1999. Application of the MultiScan™ system to assess chromosome polymorphism. Rocz. Nauk. Zoot. **26**: 9-19. (In Polish).

LADJALI K., TIXIER-BOICHARD M., CRIBIU E. P. 1995. High resolution chromosome preparation for G- and R- banding in *Gallus domesticus*. J. Hered. **86**: 136-139.

LADJALI-MOHAMMEDI K., BITGOOD J. J., TIXIER-BOICHARD M., PONCE DE LEON F. A. 1999. International System for Standardized Avian Karyotypes (ISSAK): standardized banded karyotypes of the domestic fowl (*Gallus domesticus*). Cytogenet. Cell Genet. **86**: 271-276.

MAZANOWSKI A., SMALEC E., BURZYŃSKA-RAK J. 1985. Comparison of Italian, Kuban and reciprocal crosses of geese. Zesz. Nauk. ATR Bydgoszcz Zoot. **123**: 69-79. (In Polish).

PERRY P., WOLFF S. 1974. New Giemsa method for differential staining of sister chromatids. Nature **261**: 156-158.

RABSZTYN A., JASZCZAK K., JASZCZAK J., KAPKOWSKA E. 1998. Inheritance of two morphological forms of chromosome 4 in Zatorska geese. Proc. 13th Europ. Colloquium on Cytogenetics of Domestic Animals, Budapest, Hungary. Allatenyesztes es Takarmanyozisenetics of Domestic Animals **48**: 72-74.

SCHMID M., NANDA I., GUTTENBACH M., STEINLEIN C., HOEHN M., SCHARTL M., HAAF T., WEIGEND S., FRIES R., BUERSTEDDE J. M., WIMMERS K., BURT D. W., SMITH J., A'HARA S., LAW A., GRIFFIN D. K., BUMSTEAD N., KAUFMAN J., THOMSON P. A., BURKE T., GROENEN M. A., CROOIJMANS R. P., VIGNAL A., FILLON V., MORISSON M., PITEL F., TIXIER-BOICHARD M., LADJALI-MOHAMMEDI K., HILLEL J., MÄKI-TANILA A., CHENG H. H., DELANY M. E., BURNSIDE J., MIZUNO S. 2000. First report on chicken genes and chromosomes. Cytogenet. Cell. Genet. **90**: 169-218.

SHOFFNER R. N., WANG N., LEE F., KING R., OTIS J. S. 1979. Chromosome homology between the Ross's and the Emperor goose. J. Hered. **70**: 395-400.

SILVERSIDES F. G., CRAWFORD R. D., WANG H. C. 1988. The cytogenetics of domestic geese. J. Hered. **79**: 6-8.

SŁOTA E. 1998. Chromosome polymorphism in swine. Roczniki Naukowe Zootechniki, Monografie i Rozprawy, Instytut Zootechniki PAN Kraków **7**: 1-58. (In Polish).

SUMNER A. T. 1972. A simple technique for demonstrating centromeric heterochromatin. Exp. Cell Res. **75**: 304-306.

ŚWITOŃSKI M. 1988. B chromosomes in the common fox (*Vulpes vulpes*), their structure, distribution, inheritance and significance. Rocz. AR Poznań. Rozprawy Naukowe **174**: 1-65. (In Polish).