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Elektroforetyczny rozdział na żelu poliakryloamidowym białek surowicy krwi siedmiu rodzin karpi

Electrophoretic separation of blood serum proteins on polyacryloamide gel in seven carp families

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Abstract — Electrophoretic separation of blood serum proteins on polyacryloamide gel was carried out in seven carp families.

They were found to differ from one another in the system and number of bands distinguishing the representants of particular families from one another. Differences in electrophoretic patterns are the criteria of genetic separatism of the investigated families and can be used to characterize the breeding material.

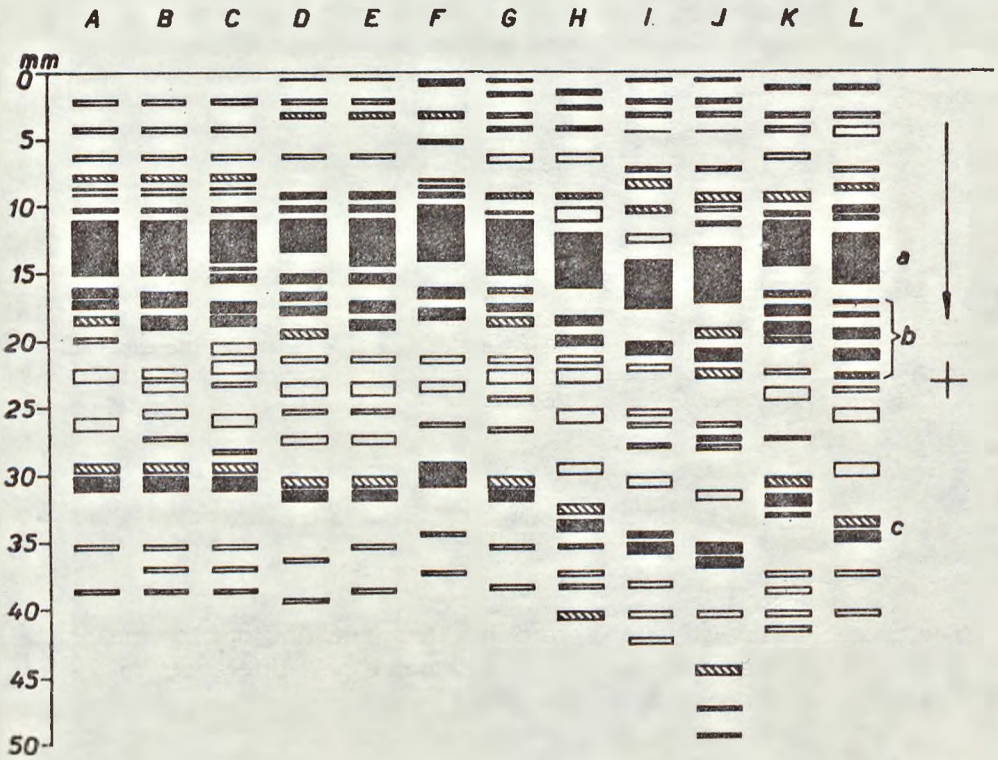
Material and method

The presented investigations were an attempt to find out, by means of electrophoretic separation on polyacryloamide gel, differences in the structure of proteins occurring in the blood serum in seven carp families, so far selected only on the basis of biometric and meristic features and on their scaliness.

The number and rate of protein band migration in the electric field depend on their structure, differentiated according to the genetic information coded in the DNA; for this reason the electrophoretic separation of proteins reflecting the protein structure is included in biochemical systematics (Thompson 1960, Tsuyuki et al. 1965, Tsuyuki, Roberts 1966, Nyman 1965, 1966, 1967, 1971, 1972, Nyman, Westin 1968, 1969, Johnson 1972, Avtalion, Prugin, Rothbard 1975, Baron 1975).

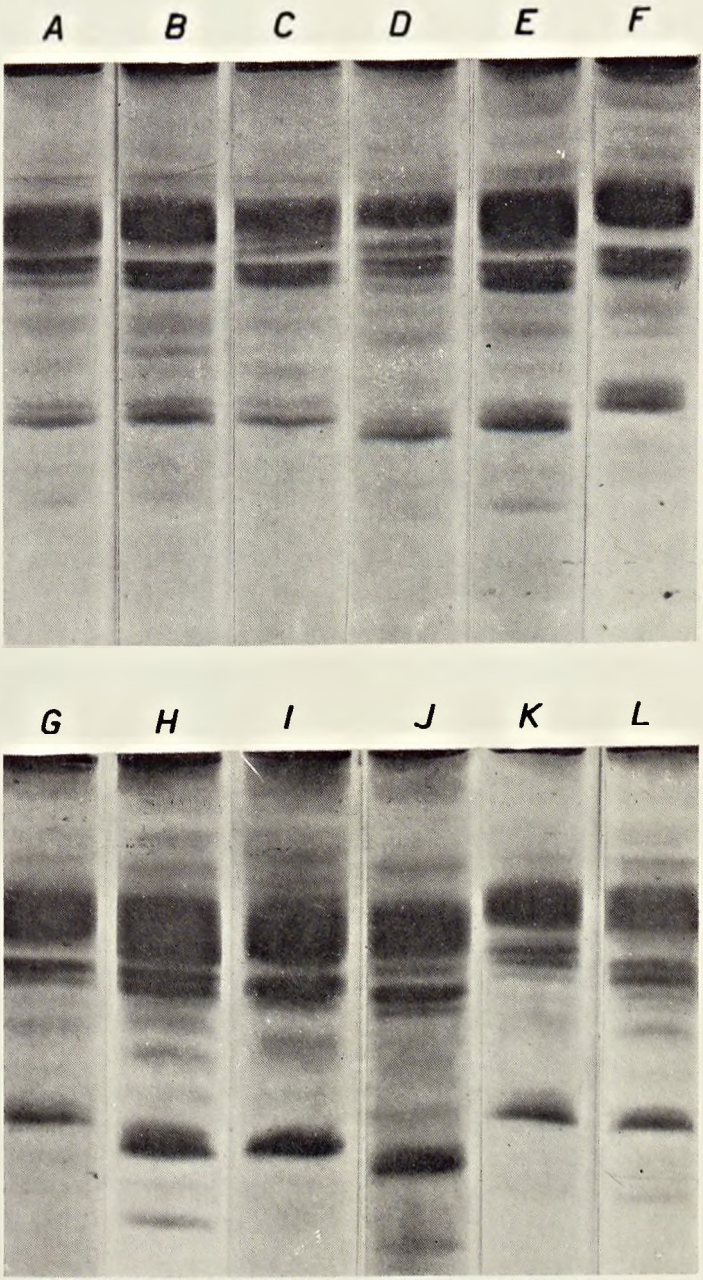
Blood serum proteins of yearlings were investigated in the following carp families: 1) the Yugoslavian thinly scaled family whose spawners were imported to Poland and placed in ponds of the carp farm at Pławowice near Cracow. These gave progeny belonging to two types of scaliness: the thinly scaled and the "leather carp". 2) Carp bred in the normal way at the farm at Pławowice and four family lines of Hungarian carp were denoted by the symbols Tölg, 77, 78, 88. Spawners of these fish were imported from Hungary to the Experimental Farm at Gołysz in 1973 and have been bred there in pure family lines.

Before taking samples of blood for investigation, the carp belonging to the above-mentioned families were placed in aquaria, where they



Ryc. 1. Schematyczne przedstawienie wzorów elektroforetycznego rozdziału białek. A, B, C — karpie jugosłowiańskie „gołe"; D, E, F — karpie jugosłowiańskie „drobnołuskie"; G — karpie pławowickie; H — karpie węgierskie „Tölg"; I, J — karpie węgierskie „78"; K — karpie węgierskie „88"; L — karpie węgierskie „77"; a, b, c — oznaczenia wyjaśnione w tekście

Fig. 1. Schematic presentation of patterns of electrophoretic protein separation. A, B, C — Yugoslavian "leather carp"; D, E, F — Yugoslavian "thinly scaled" carp; G — carp from Pławowice; H — Hungarian carp "Tölg"; I, J — Hungarian carp "78"; K — Hungarian carp "88"; L — Hungarian carp "77"; a, b, c — denotations explained in the text



Ryc. 2. Ilustracja fotograficzna wzorów elektroforetycznego rozdziału białek. A, B, C — karpie jugosłowiańskie „gołe"; D, E, F — karpie jugosłowiańskie „drobnołuskie"; G — karpie pławowickie; H — karpie węgierskie „Tölg"; I, J — karpie węgierskie „78"; K — karpie węgierskie „88"; L — karpie węgierskie „77"

Fig. 2. Photographic presentation of patterns of electrophoretic protein separation. A, B, C — Yugoslavian "leather carp"; D, E, F — Yugoslavian "thinly scaled" carp; G — carp from Pławowice; H — Hungarian carp "Tölg"; I, J — Hungarian carp "78"; K — Hungarian carp "88"; L — Hungarian carp "77"

were given similar food and kept for four weeks to permit acclimatization to identical environmental conditions. The blood serum, obtained by centrifuging the morphotic elements from blood taken from the anal vein, was stored in heparinized capillaries in deep frozen state.

The electrophoretic separation of blood serum proteins was carried out on 10 per cent polyacryloamide gel for 55 minutes at a current voltage of 2 mA per one tube 66 mm in diameter and 7.5 cm in length. For staining the protein separated on gel 1 per cent of Amido Black 10 B solution in 7 per cent glacial acetic acid was used. Staining and decoloration of the gel was carried out electrically.

Results

The patterns of electrophoretic separation of blood serum protein on polyacryloamide gel show in the investigated fish certain differences in the number and arrangement of protein bands (figs. 1, 2).

The Yugoslavian carp were represented by individuals of two types of scaliness, i.e. the "leather carp" (figs 1 and 2 A, B, C) and the thinly scaled type (figs 1 and 2 D, E, F).

Three patterns of electrophoretic separation were found in the "leather carp" for 6 investigated specimens occurring in the following relation: A — 1 : 6, B 1 : 6, C 4 : 6. The patterns differ from one another in the arrangement and number of intensively coloured bands situated between the 14th and 19th mm and light bands noticeable between the 20th and 30th mm from the start.

The thinly scaled Yugoslavian carp also had different electrophoretic patterns (figs. 1 and 2 D, E, F). Pattern D, like pattern E, was represented by two individuals. The third pattern F, was found in nine of the investigated fish. These patterns differ from one another in: 1) the absence in pattern F of the band situated in other patterns 2 mm from the start, 2) a slower by 1 mm migration of the band towards the anode in pattern F which in patterns D and E was situated at a distance of 6 mm from the start, 3) the situation of three bands between the 8th and 9th mm in pattern F instead of the two present in patterns D and E at the 9th and 10th mm, 4) a slower migration of the darker wide band towards the anode in pattern F, 5) the arrangement and number of dark bands occurring between the 15th and 19th mm, 6) a difference in the number and migration rate of the light bands between the 21st and 29th mm, and 7) a slower migration of the two bands, characteristic of all investigated carp, situated in patterns D and E between the 30th and 32nd mm from the starting point. The patterns of the "leather carp" (A B C) and of the

thinly scaled Yugoslavian carp differ from one another, above all, in the arrangement of the bands between the 1st and 10th, and the 35th and 39th mm from the start, where in the "leather carp" three and in the thinly scaled ones two bands occur and in the total number of bands which is on the average higher in the "leather carp".

The carp bred at the Experimental Farm at Pławowice showed one pattern common to all the investigated carp (figs 1, 2 G).

The group of Hungarian carp, consisting of four family lines bred in line purity (figs 1 and 2 H I J K L) do not show any variability, apart from the family "78" (figs 1 and 2 I, J) in which two patterns were observed: one (I) occurring in 22 fish out of 26 investigated ones, and the other (J) occurring in the ratio 4 : 26. Particular families, on the other hand, have different electrophoretic patterns in consequence of an unequal migration rate of particular protein bands towards the anode, causing their characteristic arrangement, and in consequence of a different number of intensively coloured protein bands situated between the 16th and 20th mm from the start.

Judging from the homogeneity of electrophoretic patterns, the patterns of carp from Pławowice and those of the families Tölg, 88, and 77 of the Hungarian group are homozygotous. In contrast, the Hungarian family 78 and the Yugoslavian family are heterozygotous, this being specially noticeable in the Yugoslavian family, whose progeny splits into two types of scaliness, "the leather one" and the "thinly scaled" one, each of them having three different patterns of electrophoretic blood serum protein separation.

In spite of common characteristic elements denoted in fig. 1 by the letters a, b, c, all the electrophoretic patterns investigated and shown in figs 1 and 2 from A to L, differ from one another in the arrangement and number of bands distinguishing the representants of particular families from one another. The arrangement and number of bands, as is well known, depends on the structure of the investigated protein, differentiated according to the genetic code. The differences in the electrophoretic patterns presented in this paper are thus a criterion of the individual genetic character of the investigated families and can be used, besides the biometric methods applied so far, to characterize the breeding material in contrast to the electrophoretic separation of the esterase group enzymes (S t a r m a c h 1977). The enzymes proved unsatisfactory as an intraspecies criterion in spite of being a good material for systematic investigations, as was proved many times by various authors.

STRESZCZENIE

Przeprowadzono elektroforetyczny rozdział na żelu poliakryloamidowym białek surowicy krwi siedmiu rodzin karpi.

Stwierdzono, że wzory elektroforetyczne pomimo wspólnych charakterystycznych

elementów różnią się jednak od siebie układem i liczbą prążków odróżniających między sobą przedstawicieli poszczególnych rodzin. Układ i liczba prążków jest jak wiadomo uzależniona od struktury badanego białka różnicowanej w myśl kodu genetycznego. Przedstawione w niniejszej pracy różnice we wzorach elektroforetycznych są więc sprawdzianem odrębności genetycznej badanych rodzin i mogą być obok stosowanych dotychczas metod biometrycznych wykorzystane do charakteryzowania materiału hodowlanego.

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