

genetics and breeding of common carp



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HYDROBIOLOGIE ET AQUACULTURE

genetics and
breeding
of common carp

VALENTIN S. KIRPITCHNIKOV

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Preface

Valentin Sergueievitch Kirpitchnikov (1908-1991) was born on August 14, 1908, at Kineshma on the middle reaches of the Volga River. He studied at Moscow State University from 1929 to 1932, and from 1932 to 1941, he worked at the Institute of experimental Biology under the direction of N. K. Koltzov. At the same time, he worked at the All-Union Research Institute of Pond Fish Culture, where he founded the first laboratory for the study of genetics and the improvement of fish production. He served in the Soviet Army from 1941 to 1945, and from 1946 to 1948 he worked at the Leningrad Institute of Biology of the Academy of Sciences. His fierce opposition to Lysenkoism, however, led to his trial before a public tribunal and the loss of his position at the Institute and prohibition of travel abroad. Supported by his family and colleagues, he was nevertheless able to continue his work at the State Research Institute of Lake and River Fisheries (GOS-NIORK) from 1948 to 1968. From 1972, until the end of his life, he worked at the Institute of Cytology of the Academy of Sciences.

Professor Kirpitchnikov published over 200 articles and several monographs, one of which has been translated in english ^[1]; he also edited several conference proceedings. His work covered a wide range of subjects, from the theoretical genetics of fish populations and the role of natural selection in microevolution, sex determination in the guppy *Poecilia reticulata*, investigations of the genetic organization of fish populations (especially of Pacific salmon), to practical studies of the genetic improvement of pond-cultured carp. His work in the latter field is particularly well-known. He was the first to develop methods for the genetic improvement of commercial fish stocks, combining selective breeding with the hybridization of different stocks. Together with his many students, he conducted three long-term breeding programmes: 1) the development of the cold- and winter-resistant Ropsha line of carp, created initially by crossing cultured Galician carp with wild-caught specimens from the Amur River, followed by eight generations of artificial selection; 2) the development of the disease-resistant Krasnodar line, by selective breeding and intercrosses of three unrelated stocks, and 3) the development of a first-generation cross between the European (Ukrainian) and wild Amur River carp, which is of considerable economic interest since it shows a heterosis effect on growth. His other investigations concerned the mode of inheritance of scale patterns in the carp and, more importantly, an evaluation of the genotypic variability of stocks

[1] Kirpitchnikov, V. S. (1981) – *Genetic bases of fish selection*. Springer-Verlag, Berlin, 410 pp.

of Pacific salmon. The latter findings were applied to an artificial breeding programme to support the commercial fishery.

Professor Kirpitchenkov was able to leave the Soviet Union for the first time in 1988, and he travelled to several countries. Together with visits to the majority of fish geneticists, he was able to visit his daughter and grandchildren in France. The crowning achievement of his long and difficult career was this direct contact with other members of the international scientific community during the last four years of his life. At the Fourth International Symposium on Genetics in Aquaculture, in Wuhan (China), he was honoured on May 21 1991, by being chosen as the first honorary member of the International Association for Genetics in Aquaculture (IAGA). On his occasion, he gave a short, concise and extremely modest speech of acceptance^[2], making no mention of the political difficulties that he had overcome in his professional work. He also gave a lecture on the selection of the Krasnodar line of carp^[3]. His last publication was a paper presented to the Belfast conference on biochemical genetics and taxonomy of fish^[4]; the manuscript of the present monograph was presented to INRA, by his family, after his death. His obituary, from which the biographical information in this preface is taken, was published by several of his colleagues^[5].

Professor Kirpitchenkov's monograph is of great theoretical and practical interest. He presents an overview of the problems of breeding carp on a commercial scale, based both on his own work, the published international literature, and a large number of important but poorly-known articles by his Russian colleagues. It contains numerous illustrations, 25 graphs and 13 tables, and integrates modern genetic techniques with a review of the classical genetic literature. His monograph will stand as the testimony of an international respected fish geneticist.

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[2] Kirpitchenkov, V. S. (1993) – Acceptance speech before the Fourth International Symposium on Genetics in Aquaculture, Wuhan, *Aquaculture*, 111: 6.

[3] Kirpitchenkov, V. S., J. Ilyasov, L. A. Shart, A. A. Vikhman, M. V. Ganchenko, L. A. Ostashevsky, V. M. Simonon, G. F. Tikhonov and V. V. Tjurin (1993) – Selection of Krasnodar common carp (*Cyprinus carpio* L.) for resistance to dropsy: principal results and prospects. *Aquaculture* 111: 7-20.

[4] Kirpitchenkov, V. S. (1992) – Adaptive nature of intrapopulation biochemical polymorphisms in fish. *Journal of Fish Biology* 40: 1-16.

[5] Schroeder, T., N. B. Cherfas, Y. P. Altukhov and G. A. E. Gall (1993) – First honourable member of IAGA, Valentin Sergueievitch Kirpitchenkov (1908-1991). *Aquaculture* 111: 3-5.

The origin of domesticated breeds of common carp

The common wild carp (*Cyprinus carpio* L.) is the ancestor of all carp breeds reared by man. Its range is vast (fig. 1). Its western part includes eastern Europe, Turkey, some fluvial basins of the Transcaucasus, the Aral Sea (until it dried up), the Syr- and Amu-Daria Rivers and their tributaries, lakes Zaisan and Issik Kul, together with some small lakes in central Asia and Kazakhstan. The eastern part of its range comprises the River Amur, numerous rivers and lakes of China and Vietnam, together with the adjacent countries of South Asia (Berg 1948; Kirpitchenkov 1967 a). It is unlikely that wild forms of the carp ever existed in Indonesia; the numerous varieties of Indonesian carp are apparently domesticated forms of the common carp brought from China or Vietnam in the 17th century or earlier (Steffens 1975).

Studies of morphological differences between European carp and Amur carp have led taxonomists to identify two subspecies: *Cyprinus carpio carpio*, found throughout Europe, the Caucasus and central Asia, and *Cyprinus carpio haematopterus*, the Far Eastern subspecies of the Amur basin and the lakes and rivers of South-Eastern China (Svetovidov 1933; Berg 1948; Nikolsky 1956). It is assumed that the divergence of these subspecies occurred towards the end of the Pleistocene, or, more probably during one of the glacial periods of the Pleistocene.

However, some investigators have claimed that the divergence between the subspecies is insignificant, "may have resulted from different ecological conditions such as temperature" (Komen 1990, pp. 6-7), and is non-hereditary, thus scarcely justifying their separation. Nevertheless, studies on the morphological features and physiological parameters of the wild carp from the two zones of distribution, together with experimental investigations of the cultivation of the two forms in identical pond conditions, show that the differences between the two subspecies are both stable and heritable. Nor can it reasonably be claimed that the differences are insignificant; in Far-Eastern carp, both the number of vertebrae and gill-rakers are considerably reduced (Kirpitchenkov 1943, 1967 a).

The wild carp of Southern Asia differs from the European and Far-Eastern subspecies, having fewer vertebrae and scales in the lateral line, and shows other morphological differences (fig. 2); it has been formerly identified as a third subspecies, *Cyprinus carpio viridiviolaceus* (Berg 1948; Kirpitchenkov 1967 a), distrib-

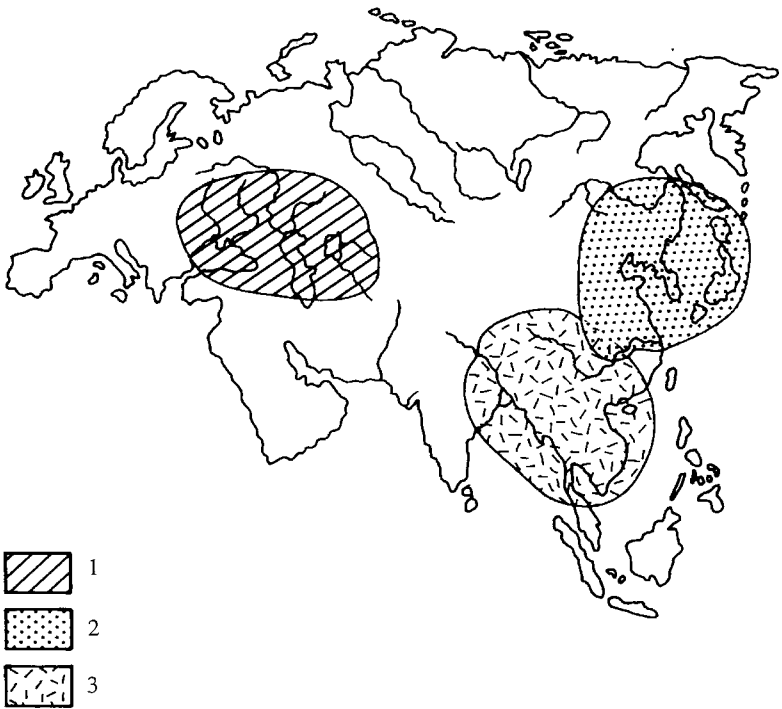


Figure 1. – Ranges of wild common carp populations in Eurasia

1. The European subspecies *Cyprinus carpio carpio*
2. The Far Eastern subspecies *Cyprinus carpio haematopterus*
3. The South-East Asian subspecies *Cyprinus carpio viridiviolaceus*

uted throughout South East China, Vietnam, Laos and Burma. However, it is highly likely that carp were introduced into Vietnam and its adjacent countries from China (Steffens 1975), and the taxonomic status of *Cyprinus carpio viridiviolaceus* is thus questionable.

Both European and Far-Eastern forms of the common carp have been domesticated for several centuries, and recently the pond culture of this fish has begun to develop in Vietnam and other Asian countries, as well as in America, Africa and Australia.

The European races of common carp go back to the wild carp of the Danube. The cultivation of wild, river-caught carp has been described by Aristoteles, and has continued throughout the Middle Ages, mainly in monastery ponds. However, the true domestication of the common carp has not begun in Europe until the

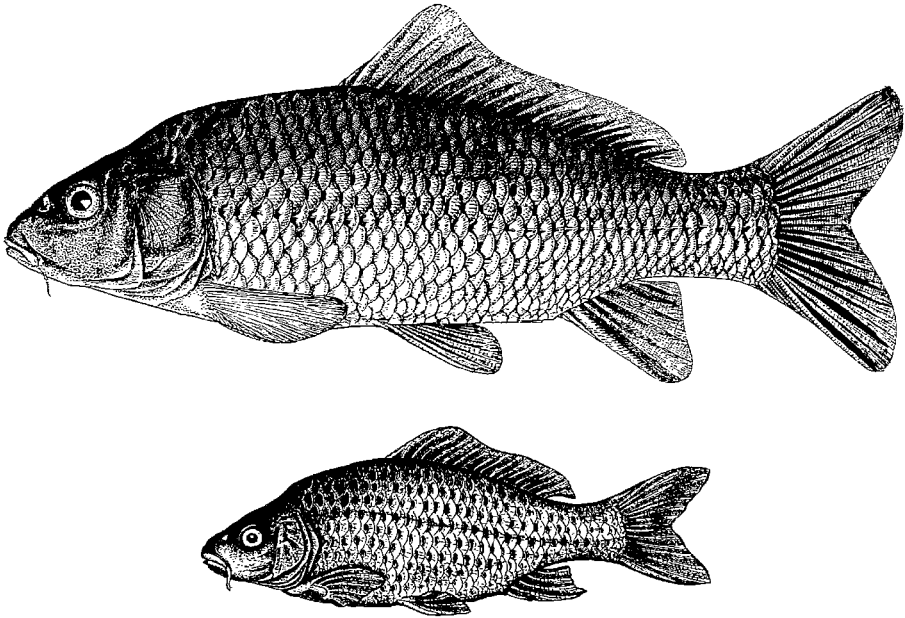


Figure 2. – Wild common carp

Above – *Cyprinus carpio carpio* from the Amur basin

Below – *Cyprinus carpio viridiviolaceus* from Vietnam (both drawn to the same scale)

16th century (Walter 1901; Wunder 1949). By the 19th century, the European cultivated carp could be divided into Galician, Bohemian, Franconian, Lausitz and Ayschgrund races (Walter 1901). Towards the beginning of the 20th century, these five races have become significantly intermingled, their characteristics changed greatly, and they have been divided into many local strains (Kirpitchnikov and Balkashina 1936). From Germany, Austria, Hungary and Poland, domesticated carp have been brought to many other European countries, including the former Soviet Union, and the other continents. Most of the strains and stocks of common carp in Germany and its neighbouring countries have disappeared during the Second World War; the development of new breeds in Eastern Europe has only begun in the 1950's.

In China, the rearing of carp in ponds has begun about 2500-3000 years ago, although the domestication began much later and was a slow process. The cultivated strains of carp in China show little difference from the wild forms; this may be due to the absence of a planned selection, as well as to the fact that larvae, fry and spawn of wild carp are often fished from rivers and mixed with the pond-reared carp (Moav *et al.* 1975 a). The Chinese carp have been transported to Japan and Korea several centuries ago, and have been domesticated in these countries.

In 1939, wild carp from the Amur basin have been first introduced into the European part of the former Soviet Union and hybridized with the European breeds to produce a number of new hybrid breeds (Kirpichnikov 1987 a; Kirpichnikov *et al.* 1993).

In Vietnam, the domestication and selection of common carp has begun in the 1950's, the hybrids between local carp and stock from Indonesia and Hungary being used for breeding. In Palestine and Israel, several European strains of common carp, as well as the cultured (Big-Belly) breed from China, have been used for the cultivation in the first half of this century. Nowadays, the common carp breeding is being increasingly undertaken in all five continents.

The karyotype of the common carp and the genetic mechanism of sex determination

The chromosome number of common carp and the problem of polyploidy

In the Cyprinidae, the diploid chromosome number is 50 in the majority of species, numbers such as 52, 48, 46 and 44 being less commonly observed. The karyotype of the common carp, on the other hand, contains about 100 elements (fig. 3); about half of these are meta- or submetacentric chromosomes, the remainder being telo- or subtelocentric. The chromosomes (Ojima and Hitotsumachi 1967; Raicu *et al.* 1972; Hafez *et al.* 1978; Zan and Song 1980; Ojima and Takai 1981). In addition, four other species of *Cyprinus* (Zan and Song 1980) and two species of Crucian carp (Ojima and Hitotsumachi 1967; Ojima *et al.* 1966, 1979; Kobayashi *et al.* 1970, 1973; Hafez *et al.* 1978; Vasilyev 1985) have the same doubled number of chromosomes.

No fewer than 10 species from the cyprinid subfamily Barbinae have also been identified as multichromosomal (Fontana *et al.* 1970; Cataudella *et al.* 1977; Hafez *et al.* 1978; Khuda-Bukhsh 1979, 1982; Suzuki and Takai 1981; Vasilyev 1985). This 17 polyploid species of cyprinid fish have been identified, and with no doubt will further investigations increase this number.

Members of the family Catostomidae, closely related to cyprinids, are also polyploid; all the species of this family studied to date have a chromosome number between 96 and 100 (Beamish and Tsuyuki 1971), and polyploidy also occurs in members of the Cobitidae (Ferris and Whitt 1977 a; Vasilyev and Vasilyeva 1982; Vasilyev 1985). Polyploidy has also been described in many other orders and families of fish; I allude here to the polyploid salmonids and whitefish, as well as to sturgeons (see Vasilyev 1985 for review).

The doubling of chromosome numbers in the common carp, Crucian carp, barbels and other multichromosomal cyprinid fish is reflected in an increased amount of nuclear DNA. In the majority of diploid cyprinids, the haploid DNA content varies between 0.7-1.4 picogrammes of DNA per cell, or from 20% to

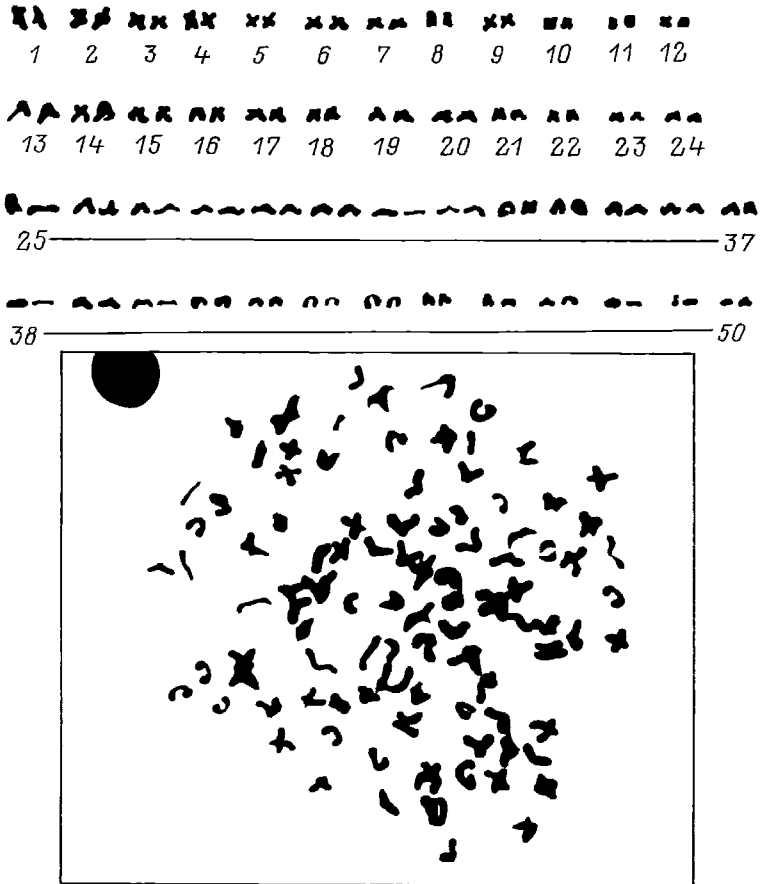


Figure 3. – Chromosomes of common carp

Above – karyotype; Below – typical metaphase plate (from Hafez *et al.*, 1978)

40% of the haploid DNA content of mammalian cells. In both common and Crucian carp, the haploid DNA content is between 1.7-2.2 picogrammes per cell, or 49%-53% of the mammalian value (Ohno *et al.* 1967; Wolf *et al.* 1969; Pedersen 1971; Ojima *et al.* 1972; see also reviews by Vasilyev 1985 and Kirpichnikov 1987 a). Polyploid cyprinids also have more erythrocytes, which are larger than those of their diploid relatives (Sezaki and Kobayashi 1978; Vasilyev 1985; Kirpichnikov 1987 a).

Polyploid fish, including carp, also possess an increased number of duplicated protein loci (Ohno *et al.* 1967; Engel *et al.* 1975; Markert *et al.* 1975; Ferris and Whitt 1977 a, b). Two (or possibly more) doublings of the chromosome set have thus taken place during the evolution of cyprinids. One of these led to the emer-

gence of the common and Crucian carp, and other members of the subfamily Cyprinae. The other (or others) have involved the ancestors of the modern large barbels, the Barbinae (Vasilyev 1985). Most investigators assign the occurrence of polyploids to the middle or second half of the Tertiary, about 40-50 million years ago (Ohno 1970; Uyeno and Smith 1972; Markert *et al.* 1975 for reviews). By way of comparison, the duplication of the karyotype in the ancestors of the modern salmonids is generally believed to be more recent, some 10-20 million years ago.

The mechanism underlying polyploidy remains unclear. Many cytogeneticists believe that the chromosome complement has doubled in hybrids between closely related species, this allopolyploidy (amphidiploidy) giving rise to the common and Crucian carp (Vasilyev 1985). On the other hand, Ohno (1970) has suggested that the evolution by autodiploidy (without hybridization) is a more plausible mechanism. Unfortunately, no evidence exists to distinguish between these hypotheses.

It is particularly striking that many polyploid cyprinids, including the common carp, barbels, and members of the genus *Tor*, are considerably larger, grow faster and are phenotypically more variable than their diploid relatives. These differences are particularly pronounced for the diploid and tetraploid members of the Barbinae (*Barbus*, *Tor*, *Puntius*). It is thus quite possible that the polyploidy confers a selective advantage, or increased fitness, associated with a larger body size, in competition with smaller, diploid forms.

The secondary diploidy has followed the reduplication of the chromosome set in the common carp. In this species, during meiosis, the chromosomes are consistently arranged in bivalent pairs, unlike the salmonids, in which tetravalents and even more complex chromosomal figures are often formed. Despite the long period of time which has passed since polyploidization, and while the common carp have approximately twice the number of ribosomal RNA genes than their diploid relatives, their cells (except for the erythrocytes) are no larger, nor do they have a higher protein content (Schmidtke and Engel 1975, 1976). The Common carp do not exhibit a larger number of nucleoli than diploid species (Cherfas 1975). The expression of genes after genome duplication thus appears to have decreased as the result of complex, unidentified regulatory mechanisms.

The karyotype of the common carp (fig. 3) consists of 48-52 meta- and sub-metacentric chromosomes, and almost the same number of subtelo- and telocentric elements (Ojima and Hitotsumachi 1967; Raicu *et al.* 1972; Hafez *et al.* 1978; Zan and Song 1980). The chromosomes are comparatively small and do not vary greatly in length.

Sex determination in the common carp

The sex chromosomes have not been cytologically identified in the common carp, although they have been recently identified in the goldfish (*Carassius auratus*) and silver Crucian carp (*Carassius auratus gibelio*) (Ojima and Takai 1979; Zan

1982). In the goldfish, as well as in many other species (but not all), the males are of the heterogametic (XY) sex, the females being homogametic (XX). Indirect evidence indicates that the common carp has the same chromosomal type of sex determination. Gynogenesis, the parthenogenetic production of diploid progeny from females, results in a uniquely female first generation of matroclinous carp (Golovinskaya *et al.* 1963 for review), a state of affairs which is only possible if the parental females are homogametic. Similar experimental studies of androgenesis have shown that the male carp possess both X and Y chromosomes (Grunina *et al.* 1990). These conclusions are also supported by data concerning the hormonal transformation; females, transformed into males by the effects of artificially administered male sex steroid hormone, transmit only X chromosomes to the offspring (Gomelsky and Cherfas 1982). It is thus almost certainly the case that the X and Y chromosomes of the common carp are of similar size and cannot be distinguished by current cytogenetical techniques. To date, no cases of X- or Y-linkage have been described in the common carp, and obviously further work in this direction is needed.

As in most fish, the Y chromosome of the common carp probably differs from the X chromosome only by one sex-determining gene or by a small block of such genes. This conjecture is supported by the finding that the male carp have been described with two Y chromosomes (Grunina *et al.* 1990). Hermaphroditism, which is quite common in carp, is also an evidence that the genetic sex determination is not very strict. It can be assumed that "male" and "female" genes are born both on the sex chromosomes and on the autosomes, and that their balance can be tipped in either direction. The functional value of unpaired Y chromosomes must also facilitate the polyploidy. New polyploids do not show the drastic perturbation of the sex ratio which is typical of species with a more strict sex determination (such as mammals and insects), and do not show reduced viability (Viktorovsky 1969; Ohno 1970). This state of affairs may possibly account for the relatively frequent cases of reduplication of the chromosome complement in the evolution of fish which I have described above.