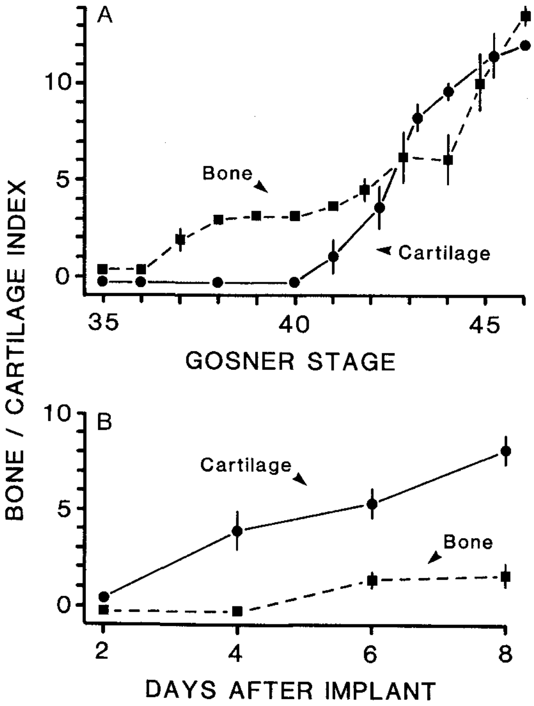

Figure 3. Relative timing of cranial cartilage and bone development. *A* During natural metamorphosis, bone formation initially precedes cartilage transformation (stages 37–40). *B* Following T_3 administration, events in the two tissues are dissociated and the natural sequence is inverted: cartilage transformation now proceeds in advance of bone formation. Ossification, but not cartilage transformation, is most advanced at this treatment regime (dosage, 2.5 µg; implant stage 32/33); the difference between bone and cartilage indexes after 8 days is even larger at lower dosages and earlier implant stages (table). Values denote means ± 2 SE.

the necessity to consider both local and systemic factors in analyses of cranial development and evolution[12].

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The role of muscle in determining growth and size in teleost fish

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Summary. Rapid growth to large size in fish results from a sustained 'recruitment' of new fibres into their axial series of myomeres. Cessation of recruitment at a small fish size leads to slow growth and a small final size of the fish. Fibre growth dynamics of fishes evidently govern growth and size through fibres' surface area to length ratios, which control their nutrient assimilation rates.

Key words. Fish muscle growth; muscle growth dynamics; fish size and growth.

It is well established that, among normal fish, growth rates can display great intraspecific variation in response to food supply, temperature[1] and, less obviously, to light and oxygen. Because of this growth 'plasticity', care should be exercised in attempting generalizations about the growth potential of a particular species. The best bases for interspecific comparisons of growth are the most rapid growths reported for different species. But it is glaringly obvious that, however evaluated, a great range of intrinsic differences exists between the growth potentials and the maximum recorded lengths (MRLs) among teleost species[1, 2].

In this article we describe the role of axial muscle in influencing both the growth of fish, and in determining their maximum attainable size.

Facts about muscle growth in fish

There is considerable evidence that the organismic growth of teleosts is limited by that of the body's predominant tissue mass – viz., muscle: the axial, bisymmetrical series of myomeres. (Note that, in all species we have studied, white glycolytic fibres comprise more than 90% of the total volume of all fibre types (white, red, pink) that constitute axial muscle[1, 3–5]. It is the preponderant white fibres to which we refer; our growth studies have not included pink and red muscle except to establish their extent by histochemical staining in the muscle systems of those species we have investigated.

As is generally understood, muscle tissue in fish becomes post-mitotic and highly differentiated following early development[1, 6]. Therefore it is unable to exhibit sustained hyperplastic growth by simple mitotic division of existing cells to produce a tissue that, no matter how large it may become is, like the liver, always composed of cells of approximately the same dimensions. Myosatellite cells are involved in the origin of fibres, and probably also in their continued production as fish grow[1, 6, 8]. This production of new fibres is characterized by the presence of fibres of < 20 µm diameter in histological sections of muscle, signifying the process we have termed 'recruitment'[1]. It appears to be an invariable rule in all species examined that recruitment of new fibres occurs early in the life of growing fish and continues for periods that show great interspecific variations. Cessation of recruitment appears final – i.e. it never recurs; subsequent increase in muscle (and fish) size result solely from the continuation of the other aspect of muscle growth – enlargement of diameter and length of existing fibres. The balance of evidence indicates the origin of fibres is largely, if not entirely, organized by the activity of myosatellite cells, though growth by fibre splitting or budding has also been claimed for eels[9].

We investigated the relative frequency distribution of fibre diameter classes (each class occupies 20 µm) in the epaxial muscle among comparable body-length classes for ten North American species of teleosts of five families, covering a great range of growth potentials and MRLs (10 to 164 cm)[2]. Among these fish, the small slow-growing species (e.g. bluntnose minnow *Pimephales notatus* and longnose dace *Rhinichthys cataractae*) cease to recruit new fibres into their muscle while they are still very small[1, 2]. However, progressively larger maximum lengths and higher potential growth rates are matched by fibre recruitment that is maintained to increasingly great fish lengths – i.e. the cessation of fibre recruitment occurs at greater fish lengths as MRLs increase. Because of sustained recruitment of new fibres (i.e. < 20 µm diameter) the mean diameter in the muscle of the very large and fast growing muskellunge *Esox masquinongy* at 70 cm fork length is no greater (i.e. 60 µm) than that in the little bluntnose minnow at 7 cm fork length. The MRL for each of the North American species plotted against fish length at which recruitment ceases ($r = 0.95$) gives a linear regression which indicates that, for these species, white fibre recruitment ceases at approximately 44% of the MRL[2].

Implications of the findings

Metabolism of tissues involves exchange processes (respiration, assimilation-excretion) at surfaces of cells (here muscle fibres). A relatively small average fibre diameter – the result of sustained recruitment of new fibres – means that there is a comparatively high average surface area per unit fibre length. The high surface area will enable processes such as amino acid assimilation and the accompanying respiration required for rapid and sustained muscle and organismic (whole fish) growth to occur at high rates. The longer that active recruitment occurs in the life of a fish the greater will be the period during which rapid growth can occur, and the greater the fish's final size (length) is likely to become.

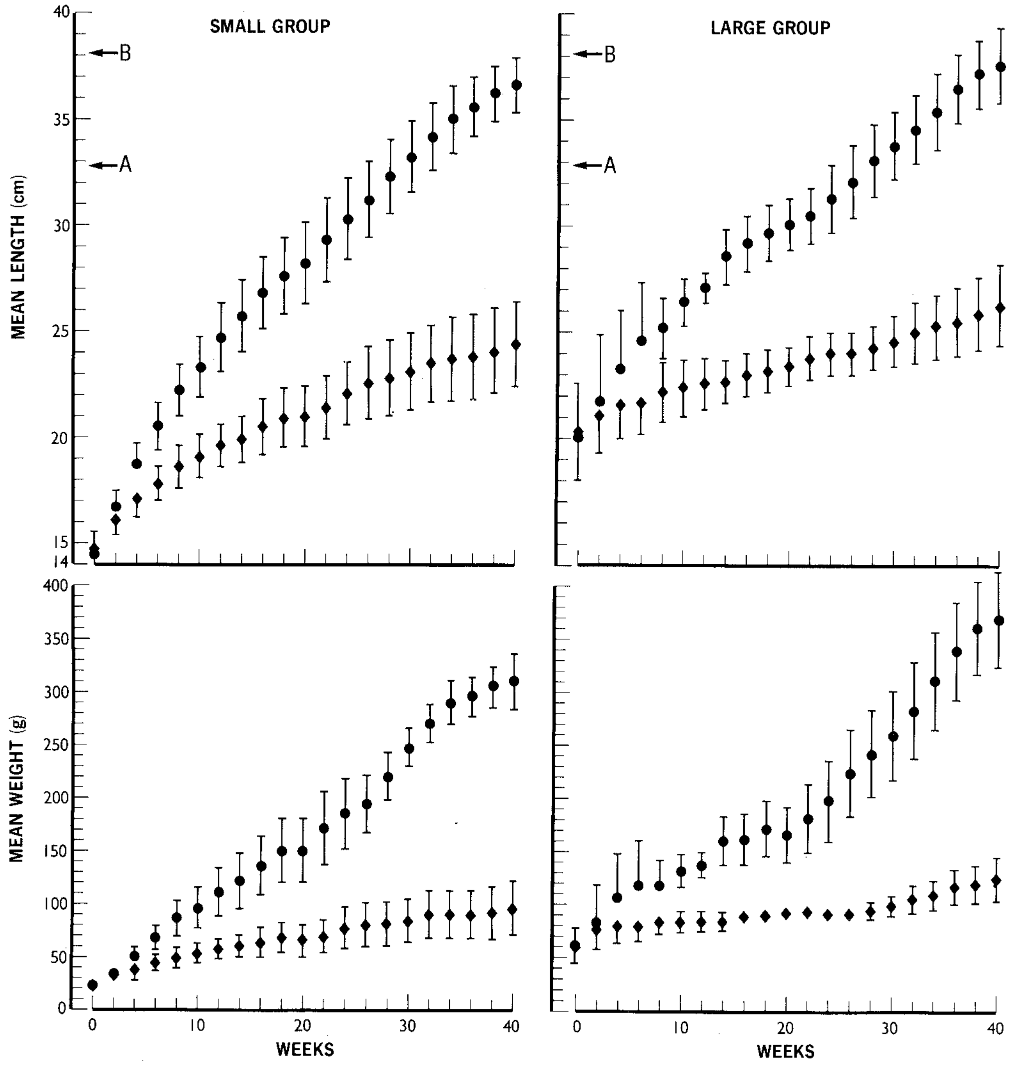
When recruitment has ceased, the subsequent growth of muscle (and fish) can result only from increase in diameter and, of course, length of existing fibres. It follows that the mean surface area per unit fibre length will decrease continuously as fibres (and fish) grow larger, producing fibre surface area to volume ratios that are increasingly unfavourable for metabolic exchanges. Hence, growth will tend to slow. There will be a limit to the value of surface area per unit fibre length, below which fibres will be unable to perform the metabolic requirements for further increase in volume (and therefore for fish organismic growth). This does not necessarily mean that a fish in which the average fibre diameter has reached the limiting value for further growth is incapable of an otherwise normal life and indefinite survival. There may be interspecific differences, but the available data point to 120–270 µm as the likely range of fibre diameter values limiting to growth[2]. It appears unlikely that tissues capable of normal hyperplastic growth resulting from mitotic divisions of cells – e.g. liver – could limit fish growth or maximum size in the same way as muscle. It is the particular growth dynamics of muscle that enable it to impose limits on its own bulk and that of whole fish.

The foregoing view of fish growth appears to offer a valid explanation for interspecific differences in the growth of muscle and, therefore, of whole fish. It can also account for the great range of maximum sizes reported among fish species. This explanation is not concerned with biochemical or molecular processes that may be highly significant in growth, but rests solely on assumptions about the metabolic consequences of simple geometrical results of muscle growth dynamics at the level of a particular tissue – axial muscle.

While we believe that our studies have indicated an important basic mechanism whereby growth potential and maximum attainable size may be determined in fish, we have not so far investigated the precise role of myosatellite cells as producers of muscle fibres, though there is evidence of their role in this regard, especially in young rainbow trout[6]. We plan to investigate the significance of their function in the later muscle growth of fish, especially as fish approach and surpass the length at which recruitment ceases for their species.

In a study of the fibre dynamics of four teleost species[10], we noted that in the grass pickerel *Esox americanus vermiculatus*, a species with relatively rather slow organismic growth and MRL of 38.1 cm[11], the fibre diameter frequencies resembled those of large and fast-growing species such as rainbow trout *Salmo gairdneri* (MRL, 103 cm) and the grass pickerel's own giant congener, the muskellunge (MRL, 164 cm). This means that even among field-collected grass pickerel as large as 25 cm there was clear evidence of sustained fibre recruitment. Therefore it was postulated that this species might be an exception to the rule that its relatively slow growth and small MRL were related to size at cessation of fibre recruitment and that it might possess the capability for much more rapid growth to a larger final size. It was proposed that a deficiency of some endocrine growth factor could be mainly responsible for its comparatively slow growth and small size.

We have performed the following experiment to investigate this proposal. Grass pickerel are reported as reach-



Curves for length and weight increase of grass pickerel in two size groups. A and B are Canadian and North American record sizes, respectively[11]. Thirteen fish receiving bGH exceeded the Canadian record size and three exceeded the North American record. Symbols for means of lengths and weight: ◆, Controls; ●, bGH-injected. Bars give 95% confidence limits. After Weatherley and Gill[13].

ing sexual maturity at the minimal size of 15.7 cm (fork length) for females and 14.1 cm for males[11]. Based on these length criteria we divided field-collected grass pickerel into two groups (each, n = 24), of respective mean total lengths 14.5 and 20.0 cm, assuming that the 'small' group represented immature and the 'large' group mature grass pickerel. At this time, the ages of fish in the experiment were estimated as around 2 years for the 'small' group and 5 years for the 'large' group, based on earlier work[12]. Each group was further divided into two subgroups (each, n = 12), all maintained in filtered, recirculated water at 22 °C and fed ad libitum on live minnows (mainly *Umbria lemi, Pimaphales* spp. and *Notropis* spp.). One subgroup of each group received bGH (bovine growth hormone) by i.m. injection (40 µg/g/2 wks). The other two (control) subgroups received i.m. injections of isotonic saline[13]. The results are illustrated in the figure[13].

Not only did the fish receiving bGH grow much faster than the controls, or grass pickerel in the field[12], their growth resembled that of their giant congener the muskellunge at a similar size[14-16]. Towards the end of the study, which slightly exceeded the 40 weeks of growth graphed in the figure, several of the grass pickerel receiving bGH exceeded the MRL for the species (see fig.). The growth of salmonids and other species of teleosts can be readily stimulated by administration of comparable doses of bGH[1]. However, the order of the growth response with grass pickerel (table) was at least 4–8 times greater than that normally reported for salmonids given bGH, indicating the action of a different type of response mechanism in grass pickerel – viz., the activation of a dormant fibre recruitment system. No other hormone was used in addition to bGH in this experiment, as the intention was simply to determine whether the latent ability to grow significantly faster and larger than normal could be elicited by hormonal stimulation.

We have also found that very small species, such as bluntnose minnow and longnose dace, in which recruitment ceases at 6–7 cm, display no response to bGH at similar dose levels to the above. Neither was there a response in bluegills *Lepomis macrochirus*, which have similar growth and MRL to grass pickerel, but in which recruitment ceases at 15 cm. Thus, in these species, failure to respond to bGH seems linked to relatively early loss of recruitment capability. In their ability to respond so amply to bGH, grass pickerel are certainly grouped with such species as rainbow trout, which can normally grow much faster and reach a much greater final size[1,2].

We have discussed the plasticity of fish growth but one further illustration of the functioning of muscle growth dynamics should be mentioned. In two experimental studies with rainbow trout[17,18], and one with bluntnose minnow[19] (i.e. a large, fast-growing, and a small, slow-growing species, respectively) the analyses of fibre dynamics indicated that fish of each species exhibited diameter frequencies characteristic of length classes that were largely, if not entirely, unaffected by even major intraspecific growth rate differences produced by differences in ration size, temperature, or the stimulating effect of bGH in the case of rainbow trout, and temperature in bluntnose minnow. We take these facts to mean that: 1) within intraspecific limits, rates of fibre recruitment and diameter enlargement are precisely linked to the requirements of organismic growth rate; 2) the mechanism of muscle growth ensures that the number of fibres and their diameter frequencies will remain linked to fish size, regardless of interspecific differences in growth rate. These are understandable principles if fish are to be ensured the requisite degree of muscular activity and swimming ability appropriate to their size and species and not to have these relationships more or less seriously disordered by differences in growth rate.

Growth rates (percent change in length and weight per day)

Treatment	'Small' group Growth rate		'Large' group Growth rate	
	Length	Weight	Length	Weight
Control	0.18	0.49	0.09	0.26
bGH (40 µg/g/2 wks)	0.33	0.91	0.22	0.65

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