blood pressure. I.v. infusion of 1 mg of the enzyme during 2 min resulted in a rapid and significant decrease in blood pressure, whereas no changes were observed with the inactivated enzyme or saline (figs 1 and 2). These results were confirmed in all rats used (total 8 rats). The activity of the enzyme (100 µg) administered to rats lasted for 90 min. The changes in blood pressure of the 8 rats were also plotted against those of angiotensinase activity in circulation in the individual rats; significant correlations between pressure drop and enzyme activity were observed for the systolic and for the mean blood pressures (fig. 3).

In the present communication, we have presented a new possibility for treating hypertension, using LAP. This method depends on the direct enzymatic destruction of angiotensin II; the hypotensive effect is very rapid (figs 1 and 2). Therefore, this method seems suitable for the emergency therapy of a hypertensive crisis caused by overactivation of the renin-angiotensin system.

Pregnancy is characterized by a marked activation of the renin-angiotensin system[13,14]; however, the blood pressure generally remains stable. Page[15] suggested that there is an elevation in the angiotensinase activity in pregnancy serum, probably due to a direct contribution of the placenta. It seems likely that angiotensinase newly appearing in pregnancy sera is involved in the regulation of plasma and tissue concentrations of angiotensin II, and thus protects women from the elevation of blood pressure. Since we previously showed that angiotensinase activity newly present in pregnancy serum could not be distinguished from placental LAP[11], it seems reasonable to suppose that placental LAP, from the point of view of the physiology of pregnancy, counteracts the hypertensive effects of renin-angiotensin release.

Although aminopeptidase M prepared from pig kidney, which is commercially available (Sigma) was also effective in lowering blood pressure as estimated by the same procedure (unpublished data), we recommend the human placental enzyme for possible human use, for immunological reasons. Furtheremore, human placenta is easily obtainable as an enzyme source.

The potential of this enzyme for treating hypertensin caused by overactivity of the renin-angiotensin system seems promising, although extensive work is required before using this method on man.

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2 We wish to express our thanks to Dr T. Suyama, Research Laboratory, Green Cross Corporation, for much assistance. We are grateful to Dr O. Suzuki (Hamamatsu University School of Medicine, Department Legal Medicine) for his critical reading of the manuscript and useful suggestions.
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Temperature dependence of the deformability of Carp *(Cyprinus carpio)* red blood cells

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Summary. Measurements were made of the deformability of the red cells of carp at different temperatures and compared with previous results obtained with another teleost fish (yellowtail) and human red cells. Changes with temperature are similar to those obtained with yellowtail, but interpretation of differences in terms of mean corpuscular volume alone are insufficient to account for the variations, which seem to be mainly due to differences in cellular deformability of the cells themselves.

Blood viscosity increases with decreasing temperature, causing a reduced fluidity of blood in vessels at low temperatures. Red blood cell deformability, which is a critical factor for blood passage through capillary vessels, might be severely affected by temperature. Recently Hughes et al.[4] studied the temperature dependence of the passage of whole blood through micropores (5 µm diameter) in a Nuclepore filter membrane using fish (yellowtail, *Seriola quinqueradiata*) and human blood. Good fluidity of fish blood through the micropores was observed throughout the temperature range studied (10–37 °C); blood passage time decreased considerably as temperature was increased from the normal 15 to 37 °C. However, when human blood was cooled, a small change observed between 37 and 20 °C was followed by a marked reduction in fluidity through the filter below 18 °C. Blood flow through the micropores was almost impossible at 10 °C but good fluidity was regained when the temperature was raised above 18 °C[5].

This difference in the effect of temperature change on fish and human blood fluidity through micropores may reflect differences in the structure of fish and human red blood cells. Further, the temperature dependence of blood fluidity through capillary vessels may be relevant to the adaptability of animals to environmental temperature fluctuations. In order to throw light on this problem, it is necessary to study blood from a wide variety of animals. This report is the beginning of such a comparative study and describes

results obtained using carp which were cannulated routinely as a part of an investigation of their responses to hypoxia at different temperatures.

Materials and methods. 8 carp kept in fresh water circulation at 15 °C were used in this study. The dorsal aorta was cannulated[6,7] and blood sampled 1-7 times from each fish without anesthesia at intervals of 1 or 2 days. During each sampling blood was taken into 1 or 2 heparinized 1 ml syringes and used for the measurements without any modifications. After 15 min incubation at each temperature 0.3 ml blood was transferred into the 0.5 ml graduated syringe and made to flow through the filter by applying 10 cm H_2O negative pressure in the reservoir connected to the outlet of the holder[4]. Complete removal of air bubbles from the filter is essential to obtain reproducible results for the blood flow[8]. The flow rate was determined for every 0.05 ml volume of the blood by measuring the times when the blood surface crossed each 0.05 ml graduation marked on the syringe. A small portion of each sample was used to determine the haematocrit (microhaematocrit centrifuge) and red blood cell number (Coulter counter). The blood flow rate through the filter, which depends on haematocrit, was converted to the mean pore passage time of single red blood cells by inserting the blood passage time, haematocrit and mean corpuscular volume of red blood cells into a previously-derived equation[9].

Results. In spite of care being taken to sample blood under as constant conditions as possible, carp blood showed large variations in its fluidity through the micropores not only between individual fish but also from one sampling to another from the same fish. In some cases, the blood flow rate through the filter decreased considerably during the passage of 0.3 ml blood. Some pores in the filter seemed to become transiently blocked by less deformable blood cells during passage of the blood. In such cases the estimation of mean pore passage time for single red blood cells becomes impossible. However, as a first approximation for those cases, the flow rate of the initial 0.05 ml blood volume was used to estimate pore passage time, since interference due to blocking of micropores would be least during that initial flow period. The mean pore passage times of single red blood cells are plotted against the mean corpuscular volume in figure 1, in which the results for human and yellowtail blood obtained in a preceding study[4] are also shown. The pore passage times of carp red blood cells through the 5 μm pores are much greater than those of human and yellowtail red blood cells probably because of their larger volume. Although absolute values for the pore passage time of carp red cells are about 10 times greater than those of yellowtail red cells, the relative changes with temperature are remarkably similar (fig. 2).

Discussion. The mean corpuscular volumes of human, yellowtail and carp red blood cells obtained in our studies are 95 ± 4, 133 ± 12 and 186 ± 24 μm³ respectively, and thus in the ratio 1:1.4:2.0.

The frictional force which acts between the surface of a red blood cell in a pore and the pore wall will increase with nearly the same ratio because the contact area increases directly with cell volume. However, the pore passage time seems to increase rather abruptly as the red cell volume exceeds 150 μm³; nor is it correlated with cell volume in a given species. These results indicate that the pore passage time of a red blood cell may be affected very little by the frictional force or lubrication factor. As suggested from the time course for the pore passage of a red blood cell schematically shown in Kikuchi et al[7], most of the pore passage time will be the time required for the red cell to be deformed to a cylindrical shape when entering a pore. This deformation time is obviously dependent on the extent of deformation and the cellular deformability.

Gregersen et al.[10] showed that the passage of a red blood cell through a pore becomes severely restricted when the cell volume exceeds a certain value, and becomes impossible unless accompanied by stretching of the cell surface area. Although the volume of a carp red blood cell is within this limit with respect to the 5 μm pore, the cellular deformation on entering a pore may cause stretching and increase in area in some parts of the cell membrane, which

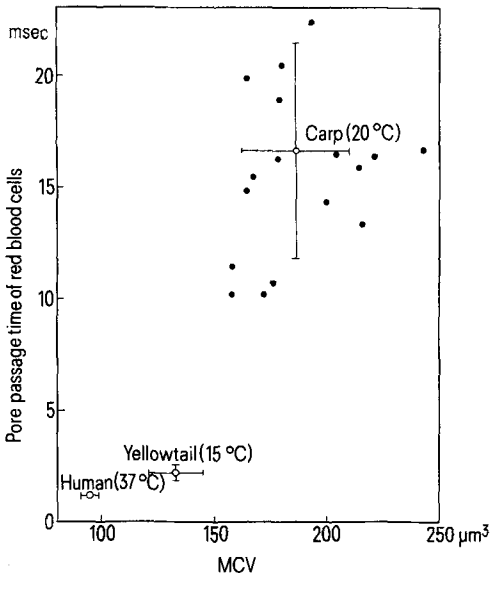

Figure 1. Relationship between pore passage time for a single red blood cell and the mean corpuscular volume (MCV) for red cells of man and 2 species of teleost fish. In each species measurements were made at the environmental temperature at which the blood was sampled. Mean values with SD are shown.

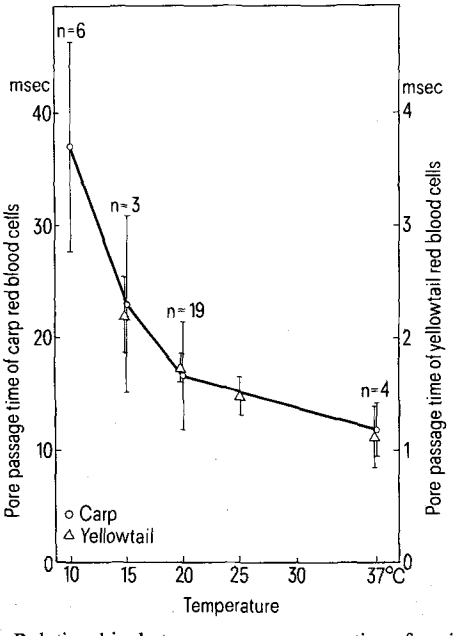

Figure 2. Relationship between pore passage time for single red blood cells of carp and yellowtail determined at different temperatures. Notice that there is a 10-fold difference in the scales for the passage time for the 2 species. For carp, SD are given together with the number of determinations.

could cause the pore passability of carp red blood cells to be much less than that of human and yellowtail red cells. From this point of view a strong dependence of pore passage time on cell volume is to be expected for carp red cells. However, as seen in figure 1, there seems to be no correlation between them. Furthermore, it was often found that repetition of blood sampling from the same fish caused large but systematic changes in the pore passability of red blood cells without there being any observable changes in mean corpuscular volume. Therefore, the large variation observed in pore passability of carp red blood cells is only partly attributable to differences in cell volume and seems mostly to be due to variability in cellular deformability itself. Loss of blood may stimulate complex responses in the fish which result in the deformability of red blood cells becoming altered considerably. Changes in the population of red blood cells having different deformabilities might also be involved in these modifications.

The pore passage times of carp red blood cells decreased with increasing temperature in a fashion similar to that observed for yellowtail red cells. The increased pore passability of red blood cells at high temperatures will be advantageous for the blood circulation, which must meet greater demands of oxygen and nutrients as a result of the elevated metabolism. The temperature dependence of the blood fluidity through micropores which was observed for carp and yellowtail seems to be consistent with the fact that both fish can live in a wide temperature range.

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On the role of carbonic anhydrase in the anticonvulsant effects of triethyltin (TET)[1]

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Summary. Triethyltin (TET) and acetazolamide (ACTZ) both produce marked anticonvulsant effects as evaluated by the maximal electroshock seizure test. However, the mechanism responsible for the anticonvulsant effect of TET and ACTZ is probably not the same since TET does not inhibit the activity of carbonic anhydrase isozyme C in vitro.

Millichap reported that the development of susceptibility to the maximal electroshock seizures (MES) test in the newborn rat is directly related to the levels of the enzyme carbonic anhydrase in the brain[3]. It was suggested that the brain carbonic anhydrase and its catalytic activity are essential for seizure discharge propagation[3]. Further studies with acetazolamide (ACTZ) suggested that low levels of enzyme activity may be sufficient for a focal discharge and the induction of a minor seizure pattern while a high degree of activity was required for a generalized seizure discharge and the induction of a major tonic seizure[4]. In addition to the increased levels of brain carbonic anhydrase, the development of MES susceptibility was related directly to a decrease in brain carbon dioxide levels, inversely to the ratio of extracellular to cellular sodium, and directly to the ratio of cellular to extracellular brain water[5].

Triethyltin (TET) is a member of the organotin class of compounds which are being increasingly used as biocides, preservatives, catalysts, and polymeric stabilizers[6]. TET produces electrolyte alterations which result in white matter edema of the brain and spinal cord, intramyelinic vacuolation and myelin splitting in the rat[7,8]. TET also delays the ontogeny of the MES patterns in developing rats[9] as well as decreases MES responsiveness in adult rats[10]. In order to determine if TET produces its anticonvulsant effect by an inhibition of brain carbonic anhydrase, this study was undertaken to compare the effects of TET and ACTZ on the MES test in adult mice and on a commercially available preparation of carbonic anhydrase C, the isozyme reported to be specific for the nervous system[11].

Outbred male albino mice (25–30 g) were obtained from Timco (Houston, Texas). 5 animals were house per cage and were provided water and Purina chow (Code No. 5008) ad libitum. Animals were maintained on a 12:12 light:dark cycle commencing at 07.00 h. Animals (19–20 mice/group) were injected (i.p.) between 08.00 and 09.00 h each day, in order to avoid circadian rhythm effects, with 0 mg/kg, 100 mg/kg or 200 mg/kg ACTZ (Lederle Laboratories, Pearl River, N.Y.) or 0 mg/kg, 1 mg/kg or 5 mg/kg TET-Br (Alpha Products, Danvers, MA). Additional groups were injected with equimolar doses of stannic bromide ($SnBr_4$), sodium bromide (NaBr) or ethanol to control for the possible ionic effects of tin or bromide or the vehicle effects of ethanol. All animals were dosed with a constant injection volume per body weight (1.0 µl/g b.wt).

All animals were seizure evaluated at 0.5, 4 and 21–24 h following injection. The methods used for eliciting and scoring the MES patterns were modified from techniques described by Woodbury and Davenport[12] and Fox et al.[13]. A 25 mA current was utilized for all mice tested. The indices measured in this study were forelimb flexion, forelimb extension, hindlimb flexion and hindlimb extension. The duration of each phase was determined and then