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INFERENCE ON THE PHYLOGENETIC RELATIONSHIP OF THE GENUS *GLYPTOCEPHALUS* (PLEURONECTIFORMES: PLEURONECTIDAE) BY SHAPE

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Tai-Sheng Chiu (1990) Inference on the phylogenetic relationship of the genus *Glyptocephalus* (Pleuronectiformes: Pleuronectidae) by shape. *Bull. Inst. Zool., Academia Sinica* **29**(2): 95-104. A set of morphometric data taken from *Glyptocephalus* and its sister genus (*Tanakius*) were used to elucidate the trait similarity among species and to infer their phylogenetic relationship.

Traditional morphometric method was based to measure a sample of 141 concerned juvenile and adult flatfishs. Each individual was then translated into a vector of 22 elements (measural variables). Within this character hyperplane, the first three principle component scores, conveyed 99% of distinctive information, were subjected to shape analysis in order to realize their phylogenetic relatioship.

The findings of shape analysis are: 1) prominent insertion of right pectoral ray is unique in *G. zachirus*, of which the structure may have high implication to its specific upwelling environment; 2) *G. stelleri* is most similar to *G. cynoglossus*; 3) cogeneric species are grouped together rather than monotypic sister, *Tanakius*; and 4) the traits are patriotic in systematic relationship, but part of it different from the conclusion made by Sakamoto (1985) with reference to inter-generic relationship.

Key words: Phylogenetic relationship, Genus *Glytocephalus*, Sister genus (*Tanakius*).

Genera *Glyptocephalus* and *Tanakius* are right-eyed flounders. Two species, *G. stelleri* and *T. kitaharae*, are distributed in the continental shelf of north-western Pacific (Masuda *et al.*, 1984). *G. cynoglossus* is distributed in the North Atlantic (Bigelow and Schroeder, 1953), while *G. zachirus* in the northeastern Pacific upwelling area (Rogers, 1985). Norman (1934) reported a close relationship between *Dexistes*, *Tanakius* and *Glyptocephalus*. Richardson (1981), based on similarity of larval morphology put *Tanakius* as sister taxon of *Glyptocephalus* logically, but believed that *Dexistes* had no relevant linkage to these genera. In consequence, she believed that *Embassichthys* should be the sister taxon of the group composed of *Tanakius* and *Glyptocephalus*. Sakamoto (1984) proposed a phenogram of Pleuronectidae which exhibited a distant relation of *Tanakius* with *Glyptocephalus* on the basis of osteology, and he also depicted a difference of generic level that *Glyptocephalus zachirus* should be read as *Errex zachirus*. Chiu (1985), based primarily on osteological study, indicated a close relationship of *Tanakius* and *Glyptocephalus* in relative to *Microstomus* and *Embathyichthys*. The present study is intended to analyze the inter-specific similarity of shape between three species of *Glyptocephalus* and one species of *Tanakius* by using traditional morphometry. The goal of this study is to clarify the interspecific similarity and to check the corroboration of difference sources.

MATERIALS AND METHODS

Materials

Among four right-eyed flounders examined, three were cogeneric (*Glyptocephalus*) and one were monotypic (*Tanakius*) which presumed to be the sister taxon of *Glyptocephalus.* Those specimens were: *G. cynoglossus* (N=70, 15.8-25.2 cm SL); *G. zachirus* (N=28; 8.03-28.7 cm SL); *G. stelleri* (N=22, 11.6-25.8 cm SL); and *T. kitaharae* (N=21, 10.5-25.8 cm SL). All specimens were preserved at museums for varying durations. Appreciation were due to MCZ, ARC, OS, LACM, HUMZ, FAKU, NSMT (institutional acronyms see Leviton *et al.*, 1985).

Measurements

In the conventional morphometric method, measurements of various body parts of representative specimens were made with calipers and/or calibrated micrometers. The measuring precision was 1 mm for measurements over 100 mm and 0.1 mm for those less than 100 mm. Measurements and their abbreviations are defined as follows (Fig. 1): 1) Standard length (SL)—snout tip to posterior margin of hypurals; 2) Head length (HL)—horizontal distance from snout tip to margin of opercle; 3) Snout length (SNL)—the shortest distance from snout tip to anterior margin of right (lower) eye; 4) Snout to preopercle length (SP)—the shortest distance from snout tip to posterior margin of preopercle; 5) Body height at anus (BH)—vertical distance from dorsal to ventral body margin at anus (excluding fin ray); 6) Left (Upper) eye diameter (LED)—horizontal diameter of migrating or migrated eye ball (eye means eyeball hereafter); 7) Right (Lower) eye diameter (RED)—horizontal diameter of non-migrating eye; 8) Interorbital space (IOS)—the narrowest space between the eyes; 9) Right maxillary length (RM)—snout tip to posterior margin of right maxillary bone; 10) Left maxillary length (LM)—snout tip to posterior margin of left maxillary bone; 11) Length of dorsal fin base (DB)—the shortest distance from the base of the first dorsal fin ray to the base of the last dorsal fin ray; 12) Length of anal fin base (AB)—the shortest distance from base of the first anal fin ray to base of last anal fin ray; 13) Snout to dorsal fin origin (SDO)—the shortest distance from snout to base of the first dorsal fin ray; 14) Snout to anal fin origin (SAO)—the shortest distance from snout to the base of the first anal fin ray; 15) Snout to right pectoral fin length (SRP)—the shortest distance from snout to the base of the first upper fin ray of right (ocular side) pectoral fin; 16) Snout to left pectoral fin length (SLP)—the shortest distance from snout to the base of the first upper fin ray of left (blind side) pectoral fin; 17) Snout to ventral fin length (SV)—the shortest distance from snout to the point in between the first upper fin ray of right and left ventral fins; 18) Length of the longest right pectoral fin ray (RPR)—direct measurement from the base of the longest right (ocular side) pectoral fin ray to its posterior end; 19) Length of the longest left pectoral fin ray (LPR)—direct measurement from base of the longest left (blind side) pectoral fin ray to its posterior end; 20) Length of the longest right ventral fin ray (RVR)—direct measurement from base of the longest right (ocular side) ventral fin ray to its

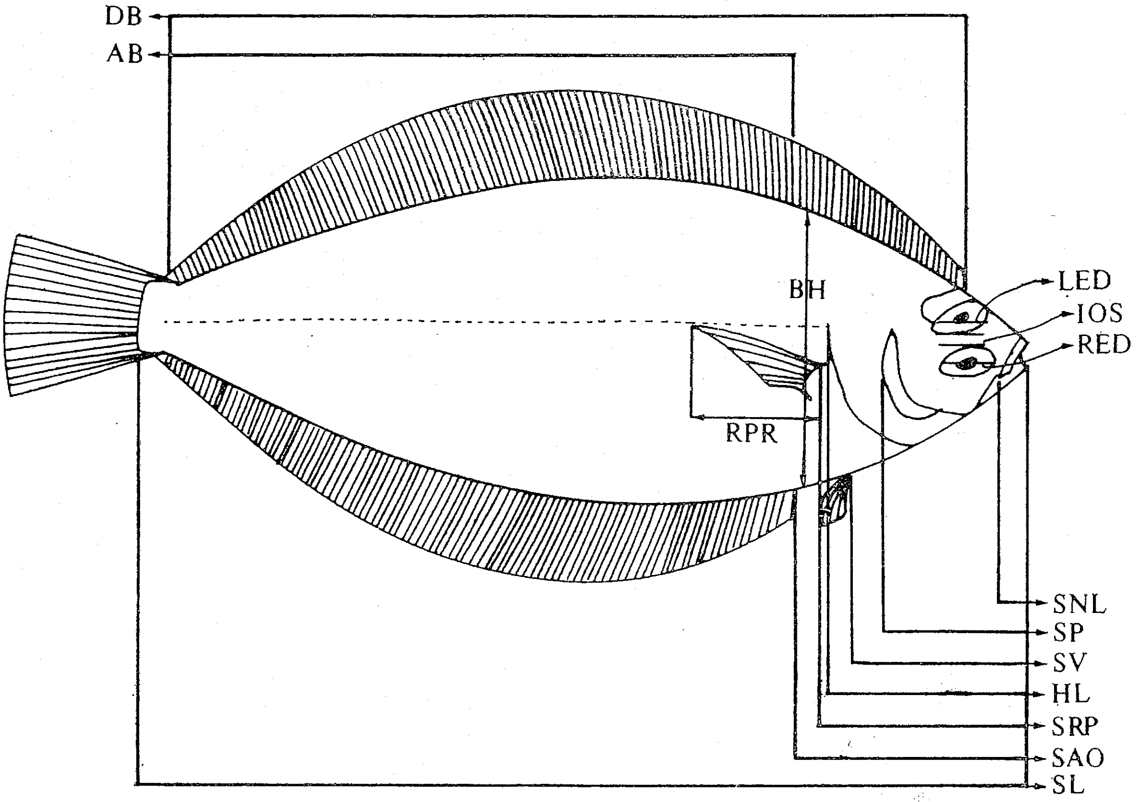

Fig. 1. A schematic diagram showing selected distance measurements. Measurements (all not included in figure) are: Standard length (SL); Head length (HL); Snout length (SNL); Snout to preopercle length (SP); Body height at anus (BH); Left (Upper) eye diameter (LED); Right (Lower) eye diameter (RED); Inter-orbital space (IOS); Right maxillary length (RM); Left maxillary length (LM); Length of dorsal fin base (DB); Length of anal fin base (AB); Snout to dorsal fin origin (SDO); Snout to anal fin origin (SAO); Snout to right pectoral fin length (SRP); Snout to left pectoral fin length (SRP); Snout to left pectoral fin length (SLP); Snout to ventral fin length (SV); Length of the longest right pectoral fin ray (RPR); Length of the longest left pectoral fin ray (LPR); Length of the longest right ventral fin ray (RVR); Length of the longest left ventral fin ray (LVR).

posterior end; 21) Length of the longest left ventral fin ray (LVR)—direct measurement from base of the longest left (blind side) ventral fin ray to its posterior end.

Statistical treatments

Dummy variable regression analysis were applied to differentiate the specific variation both on the onset of ontogenetic development (intercept) and the direction of dilation (slope). Shape score was extracted by using principle component analysis, in which the centralized second principle component was adopted to compare the patterns of variation. A logarithmic transformation of all measurements were performed for achieving data homogeneity. A cluster analysis of unweighted pair-group arithmetic average was allocated in a plane of second and third principle component.

RESULTS

Specific body allometry

An asymmetrical growth of *G. zachirus* were apparently distinguished it

Table 1
Regression analysis based on longest pectoral ray of right side vs. S. L. for identification of group differences

Variable	Coefficient	STD	t-value	p-value
const.	−0.3338	0.2949	−1.13	0.2597
X	0.1200	0.0168	7.14	0.0000
Z_1	−0.3548	0.4507	−0.79	0.4326
Z_2	0.4598	0.8424	0.55	0.5861
Z_3	0.2003	0.6494	0.31	0.7582
Z_1X	0.2122	0.0234	9.06	0.0000
Z_2X	0.0257	0.0440	0.58	0.5602
Z_3X	0.0064	0.0343	0.19	0.8513

Model: $Y=a_0+b_0X+a_1Z_1+a_2Z_2+a_3Z_3+b_1Z_1X+b_2Z_2X+b_3Z_3X+E$; for which X=S. L., Y=longest pectoral ray of right side; E=errror; Z=dummy variable for group grade; for which $Z=[Z_1\,Z_2\,Z_3]$, $Z=[0\,0\,0]$ represented *G. cynoglosssus*; $Z=[1\,0\,0]$, *G. zachirus*; $Z=[0\,1\,0]$, *G. stelleri*; and $Z=[0\,0\,1]$, *T. kitaharae*.

from the other three species, determined by following regression analysis. A dummy variable regression analysis based on the length of longest right pectoral ray, is easily used to detect the difference between *G. zachirus* and the others. The results of dummy variable analysis are tabulated in Table 1 and scatter plot is shown in Fig. 2. The separation of *G. zachirus* from the others is owing to disproportional growth of right pectoral ray with reference to standard length. This result suggests a speed-up growth of right pectoral rays on *G. zachirus* with

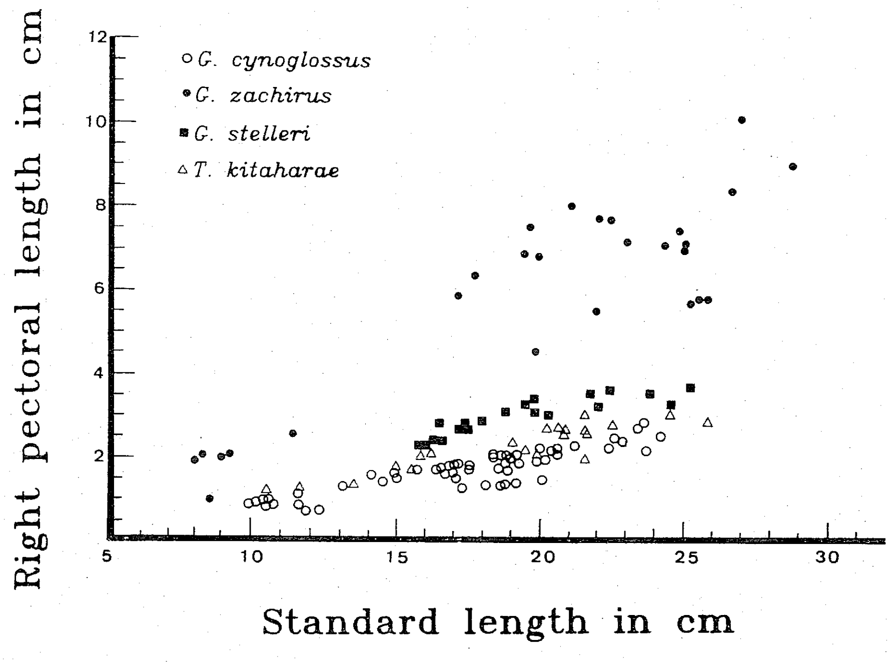

Fig. 2. Scatter of the length of longest right pectoral ray vs. standard length for *G. cynoglossus*, *G. zachirus*, *G. stelleri* and *T. kitaharae*. A discrimination between *G. zachirus* and the others could be found. The allometry between group came from differences of intercept and slope.

Table 2
Regression analysis based on longest pectoral ray of left side vs. S. L. for identification of group difference

Variable	Coefficient	STD	t-value	p-value
const.	−0.3293	0.1011	−3.26	0.0014
X	0.0873	0.0058	15.15	0.0000
Z_1	0.0694	0.1544	0.45	0.6538
Z_2	−0.0634	0.2887	−0.22	0.8266
Z_3	0.0802	0.2225	0.36	0.7191
Z_1X	0.0300	0.0080	3.74	0.0003
Z_2X	0.0287	0.0151	1.91	0.0588
Z_3X	0.0183	0.0118	1.55	0.1226

Model: $Y=a_0+b_0X+a_1Z_1+a_2Z_2+a_3Z_3+b_1Z_1X+b_2Z_2X+b_3Z_3X+E$; for which X=S. L., Y=longest pectoral ray of left side; E=error; Z=dummy variable for group grade; for which $Z=[Z_1\,Z_2\,Z_3]$, $Z=[0\,0\,0]$ represented *G. cynoglossus*; $Z=[1\,0\,0]$, *G. zachirus*; $Z=[0\,1\,0]$, *G. stelleri*; and $Z=[0\,0\,1]$, *T. kitaharae*.

a portion of more than twice longer than that in the other species. The absolute difference on coefficients of slope is 0.1766. Although the apparent magnitudes of intercept are different, the difference was not statistically significant ($p<0.05$). A dummy variable regression analysis based on the length of longest pectoral ray on left side is further tabulated in Table 2, and species coded scatter plot is shown in Fig. 3. The separation of *Z. zachirus* from the others by left side pectoral rays

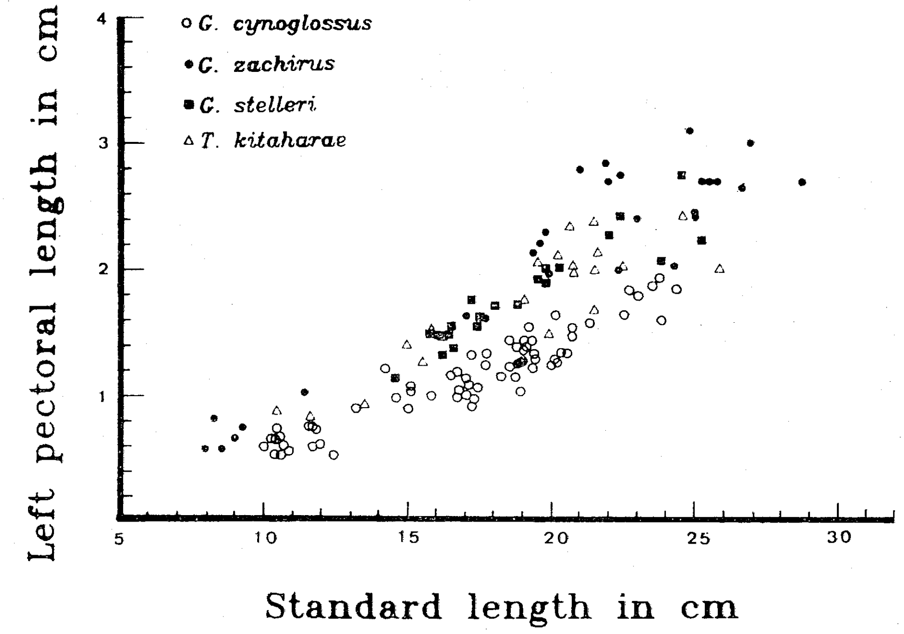

Fig. 3. Scatter of the length of longest left pectoral ray vs. standard length for *G. cynoglossus*, *G. zachirus*, *G. steleri* and *T. kitaharae*. A discrimination between *G. zachirus* and the others could be found. The difference is primarily due to early allometric growth of *G. zachirus*. At a size greater than 8 cm, relatively isometric growth could be estimated among species.

is again detectable, despite that the differences are not as obvious as that of the right side pectoral (delta =0.0087 and −0.2756 for shape and intercept, respectively).

Shape analysis

Principal component analysis based on nineteen variables was carried out for data abstraction. The first three principal component scores collected 99% of total variance. The principal component weights for first three component are tabulated on Table 3. The vectors of variable weighting on the first two principal axes are shown in Fig. 4. Standard length (SL), length of anal fin base (AB), and length of snout to dorsal fin origin (SDO) are significantly parallel to size factor, and the remaining variables are putting more contribution to shape factor (contrast). The scattering of species, coded individually, on the first two principal component axes indicated an overlapping of *G. cynoglossus* and *G. stelleri*; but a difference can be found at early stage of *G. zachirus*. *T. kitaharae* is obviously separated from *Glyptocephalus* with a minor overlapping (Fig. 5). More detailed discrimination among species can be acquired by spreading species coded individual into centralized second principal component and the third principal component (Fig. 6). These scores pull *G. cynoglossus* and *G. stelleri* apart. In summary, the shape scores based on the second principal component for determination of group and identification of inter-group relationship are sufficiently simple.

Trait similarity and relationship

The scores of the second and third principal component were applied to

Table 3
Principal component weights of the first three components. Principal component was applied to 19 morphometric variables for data abstraction. The second and third PC which may carry message of shape similarity

Variables	PC I	PC II	PC III
Standard length	.603	−.175	.127
Head length	.127	.238	−.136
Snout to opercle	.017	.053	−.028
Snout to right eye	.087	.176	−.044
Snout to preopecle	.192	.255	.108
Body height	.041	.064	−.130
Left eye diameter	.038	.044	−.094
Right eye diameter	.007	−.021	.002
Interorbital space	.531	−.209	−.563
Right maxillary length	.440	−.227	.644
Left maxillary length	.062	.092	−.077
Dorsal fin base	.166	.369	−.076
Anal fin base	.087	.176	−.045
Snout to dorsal origin	.125	.286	−.074
Snout to anal origin	.128	.454	.068
Snout to right pectoral	.128	.459	.075
Snout to left pectoral	.071	−.142	−.337
Snout to right ventral	.052	−.074	−.182
Snout to left ventral	.040	−.112	−.131

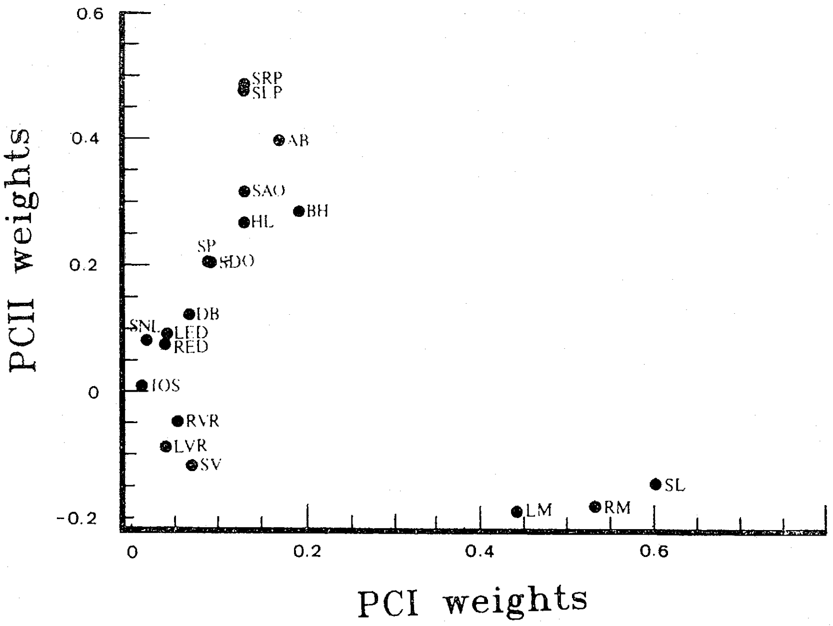

Fig. 4. Scatter plot of first two principle component weights. Standard length (SL); Head length (HL); Snout length (SNL); Snout to preopercle length (SP); Body height at anus (BH); Left (Upper) eye diameter (LED); Right (Lower) eye diameter (RED); Interorbital space (IOS); Right maxillary length (RM); Left maxillary length (LM); Length of anal fin base (AB); Snout to dorsal fin origin (SDO); Snout to anal fin origin (SAO); Snout to right pectoral fin length (SRP); Sout to left pectoral fin length (SLP); Snout to ventral fin length (SV); Length of the longest right ventral fin ray (RVR); Length of the longest left ventral fin ray (LVR).

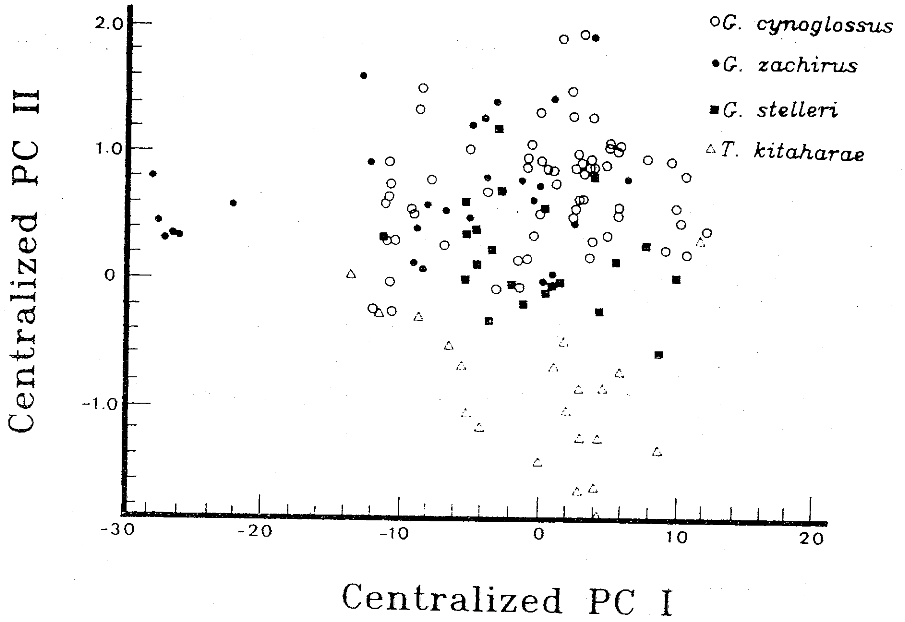

Fig. 5. Scatter plot of the first two centralized principle component scores for four flounder species, *G. cynoglossus*, *G. zachirus*, *G. stelleri* and *T. kitaharae*. A discrimination among *G. zachirus*, *T. kitaharae* and *G. cynoglossus*+*G. stelleri* could easily be found.

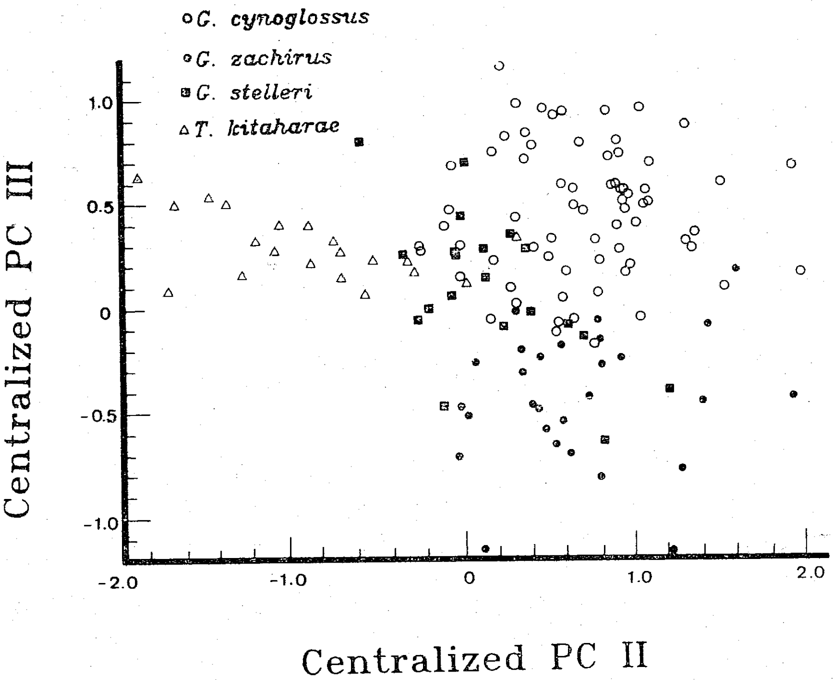

Fig. 6. Scatter plot of the second and third centralized principle component scores for four flounder species, *G. cynoglossus*, *G. zachirus*, *G. stelleri* and *T. kitaharae*. A discrimination among *G. zachirus*, *T. kitaharae* and *G. cynoglossus*+*G. stelleri* could early be found

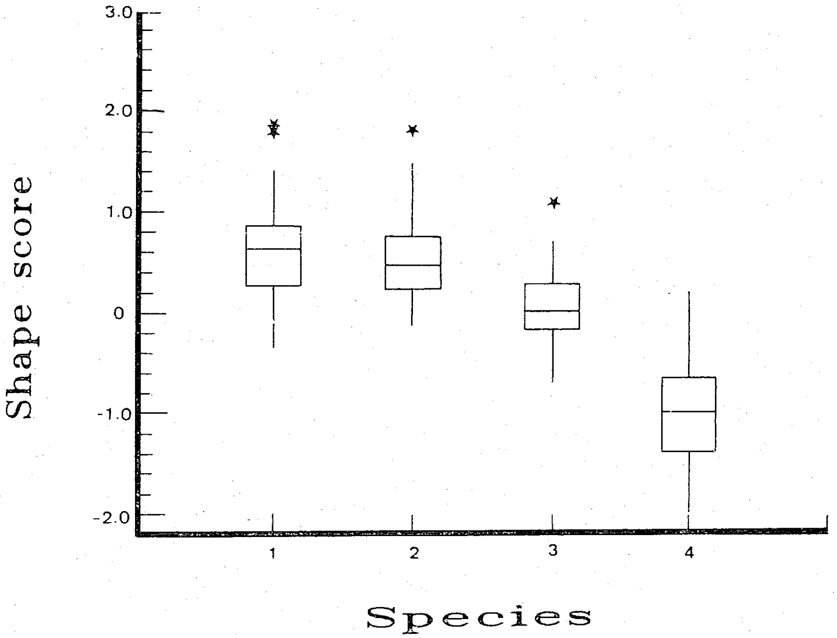

Fig. 7. Box-and-Wisker plot for shape scores of four right eye flounders, *G. cynoglossus*, *G. zachirus*, *G. stelleri* and *T. kitaharae*. A discrimination between genus *Glyptocephalus* and *Tanakius* could be found.

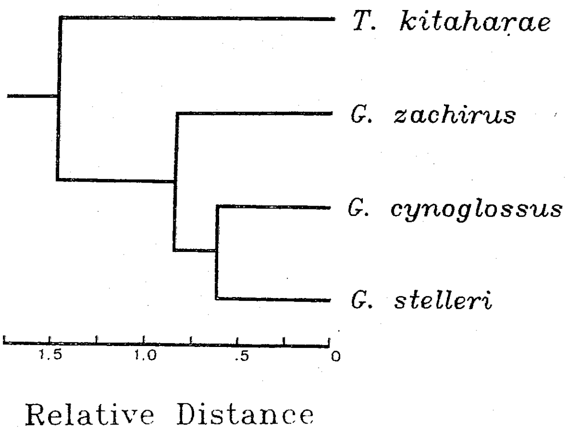

Fig. 8. A hierarchiial phenogram of four right eye flounders. A higher level discrimination could be first achieved at genus level. A second discrimination within genus was depicted by aberration of *G. zachirus* from *G. cynoglossus* and *G. stelleri*.

discriminate the traits of different species. A result from clustering method of UPGMA is translated into hierarchical phenogram as shown in Fig. 7. *G. cynoglossus* and *G. stelleri* form the first degree intimacy, while their cogeneric species of *G. zachirus* joining next. Finally, *T. kitaharae*, becomes the sister taxon of the above species group. Therefore, shape scores portray the systematic relationship patriotically.

DISCUSSION

Both Sakamoto (1984) and Chiu (1985) have supported that *G. stelleri* and *G. cynoglossus* are closest relatives. Geographically, these two species are distant —one occurs in western North Pacific and the other in North Atlantic. From the view point of intra-generic relationship an earlier character displacement occurred between *G. zachirus* and the group including *G. stelleri* and *G. cynoglossus*, the first species adapted a special environment of upwelling system can probably be inferred. Since *Glyptocephalus* does not spread further south to the Panama isthmus, a faunal exchange across arctic sea may probably quite recent. *T. kitaharae*, believed to be a sister group of *Glyptocephalus*, belongs to southern species. It's separation from its relatives may take about twice longer time than the separation of *G. zachirus* based on a hypothesis that character displacement proportional to the time elapsed since geographic separation began. Therefore, the end-products of comparative osteology, shape analysis and zoogeography have a parallel trend pointing to a clear picture of *Glyptocephalus*' speciation.

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以形貌分析推論刻首屬 (*Glyptocephalus*) 的種源關係

丘 臺 生

著者利用一組得自刻首屬（*Glyptocephalus*）的形態測定資料，來斷明它們種間的外貌相似性，進而以相似性推論它們的種源關係。

測量的方法用傳統形態學的測量法。標本數爲141尾稚魚或成魚，經測量其外部形態之後，每條魚均以此22測量值所構成的向量爲其代表。這些向量經主成分分析，將變異之99％集中在前三個主成分軸後，再以羣集分析法鑑別它們種間的親疏關係。

這些得自形貌分析的結果是：1) *G. zachirus* 的右胸鰭特別延長，可能與其適應獨特的湧昇流環境有關；2) *G. stelleri* 與 *G. cynoglossus* 親緣最相近； 3) 同一屬的種在形貌分析中最爲相似，其次才與其姊妹羣的 *Tanakius* 連結在一起；4) 形貌分析的結果能忠實地反應由骨骼學所得的推論，以及它們長久以來在系統學中的關係。