

## Protein Nutritional Quality of Precooked Seafood as Predicted by the C-PER Assay

Hong-Soo Ryu and Kang-Ho Lee\*

Department of Nutrition and Food Science, National Fisheries University of Pusan  
\*Department of Food Science and Technology, National Fisheries University of Pusan  
(Received August 21, 1984)

### C-PER方法을 이용한 水産蛋白質 品質의 豫測

柳 洪 秀 · 李 康 鎬\*

釜山水産大學 食品營養學科 \*釜山水産大學 食品工學科  
(1984년 8월 21일 접수)

#### 요 약

최대의 *in vitro* digestibility를 나타내는 조건에서 가열처리한 오징어, 굴, 새우, 명태 및 김을 대상으로 rat-PER, NPR 및 *in vivo* apparent digestibility를 측정된 결과와 최근 개발된 computed PER(C-PER) 및 discriminant computed PER(DC-PER) technique을 이용하여 계산된 결과를 비교하여, 수산단백질의 품질을 신속하게 측정할 수 있는 가능성을 검토하였다.

오징어, 새우, 명태의 C-PER은 rat-PER 보다 낮게 계산되었으나, 김은 높게 계산되었으며, 굴은 거의 비슷하게 계산되었다. 또한 이들 시료의 DC-PER은 rat-PER 및 NPR에 C-PER보다 근접된 결과를 보였으며, 단백질효율비(PER)와 소화율이 높은 것으로 알려진 굴은 가공저장 중 발생된 산패의 결과로 rat-PER 및 C-PER이 표준단백질인 ANRC casein 보다 훨씬 낮게 계산되었으나, DC-PER은 높게 계산되었다.

*In vivo* 및 *in vitro* digestibility가 높은 시료의 단백질 품질평가는 DC-PER이 정확하였으며, 품질변화가 심하게 일어난 시료나, 소화율이 낮은 시료는 C-PER technique이 유리할 것으로 생각되었다.

#### Introduction

Since seafoods can be expected to help considerably in correcting the state of malnutrition so widely prevalent in the world, it is important to develop processes for the most efficient utilization of seafoods and yet protect their nutritive value during processing. Seafoods used as samples in this study were squid, oyster, shrimp, pollock and

laver(*Porphyra tenera*) which are the popular seafoods in Far-Eastern dishes especially in Korea and Japan. These seafoods(squid, pollock and laver) are processed into dry products, marketed as raw fish(squid, oyster, pollock and shrimp) or refrigerated (squid, oyster and pollock) and processed into various types of foods such as salted(squid), smoked (squid and oyster), paste(pollock), canned(oyster and shrimp) or fermented fish sauce(squid, shrimp

and oyster).

To assess protein nutritional quality, some investigators have determined the nutritive value of seafood, mainly fish and shellfish, using amino acid analysis or animal bioassays, and they concluded the protein quality of seafoods could be compared favorably with that of meat, milk and eggs. Problems associated with the evaluation of protein quality using animal experiments include time, cost, and non-reproductibility of experiments. Therefore, low cost and rapid assay are needed to assess protein quality. Any methods used must yield data which compares favorably with conventional rat based PERs or other accepted bioassays. Recently, computed PER (C-PER) and discriminant computed PER (DC-PER) techniques using amino acid profiles and protein digestibility data have shown promise in rapidly assessing protein quality.

The present study was designed to compare computed PER(C-PER), discriminant computed PER (DC-PER) values and *in vitro* protein digestibility to rat based *in vivo* protein digestibility for precooked seafood samples treated under various processing conditions as reported by Ryu and Lee (1985).<sup>1)</sup>

## Materials and Methods

### 1. Sample Treatments

Frozen squid (*Loligo vulgaris*) and oyster (*Ostrea gigas*) were obtained from a local Asian food store. Frozen pollock (*Gadus virens*) fillets were purchased retail outlet. Peeled/deveined frozen salad shrimp (*Pandalus jordani*) were purchased locally while the sundried lavers (*Porphyra tenera*) were from Korea. All samples were kept in a  $-20^{\circ}\text{C}$  air-blast freezer until ready for preparation. The conditions of thawing and processing were the same as described by Ryu and Lee (1985).<sup>1)</sup> Thawed and heat treated samples were freeze dried for 24~26 hours at 0.5~0.75 mm Hg.

### 2. Proximate Analyses

Crude protein, crude fat, moisture, and crude ash were done on freeze samples using AOAC

(1975)<sup>2)</sup> methods.

### 3. Rat Bioassay

Rat diet preparation, required 700 grams of freeze dried squid, shrimp and pollock, and 1,300 grams of freeze dried oyster and laver. All samples were ground using a Willey Mill (model No. 3) to pass through a 80 mesh screen and then stored at  $5^{\circ}\text{C}$  until ready for mixing with other diet components. Diets were formulated using the procedure for PER as outlined by AOAC (1975).<sup>2)</sup> ANRC casein was the reference protein. Each seafood sample, along with casein, was fed to 10 male weanling rats (Sprague-Dawley strain, purchased from Holtzman Co., Madison, WI). The 21~22 day old albino rats were randomly assigned by weight to individual cages. Food and water supplied *ad libitum*. Prior to feeding the experimental diet, the rats were placed on an adaptation diet of Lab-blox 8604-00 (Wayne Allied Mills Inc., Chicago, Ill) for a 3-day period. The animal room was maintained at  $22\sim24^{\circ}\text{C}$  and 50~60% relative humidity throughout the study. Food consumption and fecal output data were recorded daily for 8 days (days through 18) of the 28 day study in order to determine the *in vivo* protein digestibility. The *in vivo* apparent protein digestibility was calculated as follows:

$$\% \text{ in vivo apparent dig.} = \frac{\text{N. in diet(g)} - \text{N. in feces(g)}}{\text{N. in diet(g)}} \times 100$$

Dunlap et al. (1974)<sup>3)</sup>

A net protein ratio (NPR) assay, which has the advantage of considering protein maintenance requirements in addition to growth requirements, was run according to the procedure Bender and Doell (1957).<sup>4)</sup> To estimate the maintenance requirements, a group of rats was fed a nonprotein diet for 10 days. The weight loss of this group was added to the weight gain of the test groups, thereby taking into account the maintenance as well as the growth requirement of the rat. The following equations were used to calculate the NPR and PER values.

$$\text{NPR} = \frac{\text{Weight gain(g)} + \text{weight loss(g) of N.G.}}{\text{Total protein consumed}}$$

N.G: nonprotein group

Bender and Doell (1957)<sup>4)</sup>

$$\text{PER} = \frac{\text{Gain in body weight(g)}}{\text{Protein intake(g)}}$$

Osborne et al. (1919)<sup>5)</sup>

#### 4. *In vitro* Assay

The *in vitro* protein digestibility of samples and casein was measured using the multi-enzyme automatic recording techniques described in AOAC (1982).<sup>6)</sup> Amino acid profiles for the samples was determined using a Beckman 120C amino acid analyzer. The samples were hydrolyzed with 6 N HCl, under vacuum, for 24 hours at 110°C to release the acidic, neutral and basic amino acids. Tryptophan was released using an alkaline hydrolysis<sup>7)</sup>, the sulfur-containing amino acids were quantitatively released using a performic acid pre-treatment of the samples followed by a 6 N HCl hydrolysis.<sup>8)</sup> The C-PER and DC-PER values for seafood samples were calculated using the procedures outlined by AOAC (1982).<sup>9)</sup>

### Results and Discussion

The proximate composition of the freeze dried seafoods are shown in Table 1. The high crude fat level(11.95%) in freeze dried squid was most likely due to the inclusion of pigments in the incompletely skinned arms of the squid. Most of the solids present were crude protein(85.23%). The composition of the squid was comparable to the spray dried squid evaluated by Lee et al. (1974)<sup>9)</sup>, and Suyama et al. (1981).<sup>10)</sup> Discoloration can occur in canned and dried oyster products from the formation of brown pigments by the condens-

**Table 1. Proximate composition (wet weight basis) of precooked and freeze dried seafood samples**

Sample	Protein (%)	Fat (%)	Moisture (%)	Ash (%)
Squid	85.23	11.96	1.40	2.04
Oyster	46.32	9.10	5.15	3.25
Shrimp	84.62	3.18	1.00	6.07
Pollock	92.41	0.76	4.03	3.83
Laver <sup>a</sup>	39.16	1.19	7.60	9.14

<sup>a</sup> Sun dried product (*Porpyra tenera*) from Korea

ation of glucose with proteins and free amino

groups. The development of rancidity associated with unsaturated fat oxidation is also a serious problem. Since these problems are derived from the characteristic component of the oyster, the determination of the proximate composition of the oyster is an important factor in nutritional evaluation. The proximate composition of the oyster varies with season, particularly with the spawning time.<sup>11)</sup> Crude fat, crude protein and glycogen generally build up during spawning, after which they tend to drop. Hatanaka(1940)<sup>12)</sup> reported the seasonal variation in whole oyster crude fat content ranged from 11.02% to 15.95% and crude protein content closely followed the same pattern. The lower fat content of the oyster used in this study, in comparison with that used in previous studies, indicated that the oysters used in this experiment were harvested during the fall season. The crude protein content of freeze dried oyster was approximately 50% which was comparable with the results of other researchers.<sup>11-14)</sup> Crude protein content in peeled/deveined shrimp was somewhat reduced, about 5~7%, from values reported by Shrinivas et al. (1974)<sup>15)</sup> and Toma and James(1975)<sup>16)</sup>, due to the losses in soluble nitrogen during the commercial peeling process. The crude fat, and moisture values for shrimp were all within the ranges reported by Shrinivas et al. (1974)<sup>15)</sup> and Gordon and Roberts (1977).<sup>13)</sup> Proximate composition of pollock was different from that obtained by Lee and Kim (1979)<sup>17)</sup> and Mihara et al. (1977).<sup>14)</sup> The differences could be due to either a difference in sampling site or the species studied. However, the results were almost the same as the data on pollock FPC from Iwaya and Yamaguchi(1979)<sup>18)</sup> and cod fillet from Power(1964).<sup>19)</sup> Mihara et al. (1977)<sup>14)</sup> stated that the protein content of laver is higher than that of the other seaweed ranging from 33~40%. The laver used in this study had a high protein content(42.38%). Its proximate composition was similar to others previously described.<sup>14, 20, 21)</sup>

The growth and protein efficiency ratio(PER) data for rats fed the seafood diets are shown in Table 2. Results from this study indicated that the

Table 2. Weight gain and protein efficiency ratio (PER) for rats fed seafood samples

Diet	Total feed consumed (g)	Total protein consumed (g)	Total weight gain (g)	Uncorrected PER	Corrected PER
Casein(ANRC) <sup>a, b</sup>	412.2±47.7 <sup>c</sup>	42.9±5.0	133.4±16.7	3.1±0.1	2.5
Squid	484.6±35.7	49.2±3.6	177.2±20.3	3.6±0.1	2.9
Oyster	318.8±30.2	32.5±3.1	77.2±13.7	2.4±0.3	1.9
Shrimp	494.8±57.4	50.1±5.8	182.4±35.4	3.6±0.4	2.9
Pollock	462.1±59.7	46.0±6.0	174.6±27.7	3.8±0.2	3.0
Laver	316.8±33.9	32.0±3.4	66.4±12.9	2.1±0.3	1.7

<sup>a</sup> Animal Nutrition Research Council<sup>b</sup> Average of 10 rats/treatment<sup>c</sup> Standard deviation

PER values for squid, oyster, shrimp, pollock, and laver were 2.9, 1.9, 3.0 and 1.7 respectively. These values compare favorably with the casein reference of 2.5. Results of a PER study on squid (2.66~3.08) by Lee et al. (1974)<sup>9</sup> showed close similarity to PER datum obtained in this study. Other authors<sup>10, 22~24</sup>) reported that squid protein was superior to casein when measured by rat assay. The oyster known to be one of the marine proteins best suited to man since its protein quality is superior to that of other seafoods.<sup>25</sup>) But in the present study, negative values were obtained in some rats fed oyster diet. From the beginning of the rat study, all oyster-diet rats experienced a considerable amount of diarrhea, and the diets smelled "stale" or of "rancid fish" odors, which was believed to be associated with unsaturated fat oxidation. Similar results for precooked oyster were obtained by Schwartz and Watts(1957).<sup>26</sup>) Because all of the protein sources used in the rat diet were stored for 3 months at 5°C after heat treatment before lyophilization, fat oxidation occurred in the oyster diet during the storage period. The resulting rancidity may have caused diarrhea and lowered feed consumption thereby reducing digestibility and weight gain. Similar results were obtained by Jones (1926)<sup>27</sup>), using not-fat extracted oyster. He reported the PER for oyster to be 1.3, 2.2 for shrimp, and 2.1 for clam. Good nutritive value obtained by Lanham and Lemons(1938)<sup>28</sup>) is most likely due to the usage of defatted oyster. The PER value of shrimp was higher than that of casein as reported in data from Matsuno(1973).<sup>28</sup>) However, Sidhu et

al. (1970)<sup>29</sup>) found that shrimp PER was slightly lower than that of casein. The pollock diet showed the highest PER value among the seafood samples used in this study. Iwaya and Yamaguchi(1979)<sup>18</sup>) also found a PER of 3.3 for pollock FPC, while Matsuno and Iwaya(1971)<sup>24</sup>) reported the NPU value of pollock to be 87.7 and 75.1 for casein. Some authors have reported higher PER values for pollock and cod. The protein nutritive value of pollock is superior to that of casein, as evidenced by significantly greater weight gains and PER values shown in this and the previous studies.<sup>18, 24, 30</sup>) The most widely known edible type of red seaweed is laver (*Porphyra tenera*), and it is used as a food source for Far-Eastern people. The nitrogen content and amino acid composition of this seaweed in general compares favorably with vegetable proteins of fairly good nutritional quality, and therefore could be expected to complement vegetable proteins of poor quality in dishes prepared for human consumption. The nitrogenous constituents of seaweed, even of laver, are known to be poorly digested by animals. This poor digestibility is thought to be due to the very tough cell walls which prevent the utilization of proteins in the cells by the digestive system.<sup>31</sup>) As shown in Table 2, finely ground(80~100 mesh) laver in rat diets shows a PER of 1.7, which was the quality shown for chlorella by Lubitz(1963).<sup>32</sup>) If the polysaccharide structure of the cell wall could be disrupted, enzyme digestion would not be hindered and the protein efficiency ratio could be increased.

To assess the nutritional quality of precooked

**Table 3. Net protein ratio(NPR) values for the precooked seafoods**

Diet	Total feed consumed (g)	Total protein consumed (g)	Total weight gain (g)	NPR
Casein(ANRC) <sup>a, b</sup>	111.5±10.7 <sup>c</sup>	11.6±1.1	34.9±5.4	4.8±0.4
Squid	124.8±11.6	12.7±1.2	53.5±7.2	5.8±0.3
Oyster	88.9±12.0	9.1±1.2	18.0±9.6	4.2±0.6
Shrimp	138.4±18.4	14.0±1.9	58.5±10.8	5.6±0.3
Pollock	129.0±14.6	12.9±1.5	54.7±10.1	5.8±0.4
Laver	87.2±10.3	8.8±1.0	13.9±3.8	4.0±0.4
Non-protein diet			-20.6±2.75	

<sup>a</sup> Animal Nutrition Research Council<sup>b</sup> Average of 10 weanling rats/treatment<sup>c</sup> Standard deviation

seafood, a net protein ratio(NPR) study was conducted. Results of the NPR study compared favorably with the PER study(Table 3). There were large differences in the consumption levels and NPR values for various diets. The consumption levels and NPR values of the oyster and laver diets were low, when compared to that for casein, pollock, squid and shrimp diets. The rancid flavor and odor of the oyster diets resulted in lowered diet consumption levels and a considerable amount of diarrhea. Morrison et al.(1962)<sup>30)</sup> reported that PER and NPR values were significantly influenced by the process and portion of fish used in preparing the diet, and by the level of dietary protein. In a one week NPR study, they obtained a 4.74 NPR value(11.5% protein level) for cod fillet diets. The NPR value of the pollock diet obtained in the present study was higher than the result obtained by Morrison et al.(1962).<sup>30)</sup> This difference was thought to be due to the different study period, the kind of samples used and the protein level in the diets. However, the results of the present study were similar to the NPR value(5.9) obtained

by Iwaya and Yamaguchi(1979) for pollock FPC diets. The NPR values for squid, shrimp, and pollock diets is significantly higher than that for casein<sup>33)</sup> and egg.<sup>34)</sup> In an attempt to determine if a relationship existed between diet consumption patterns and the PER and NPR values, a simple regression analysis was performed. The correlation coefficient for NPR values and grams of protein consumed was  $r=0.951$ ; the correlation coefficient between the NPR value and PER value was  $r=0.969$ . This analysis showed two things:1) NPR can be substituted for PER in evaluating seafood protein quality and 2) NPR values were probably dependent upon food consumption. These findings contradict data reported by Bender and Doell(1957)<sup>4)</sup> and McLaughlin(1979).<sup>35)</sup> If the rats fed the protein diets, could have been induced to consume larger quantities of diet, similar to quantities consumed by the casein diet rats, the NPR values of the protein diets would likely have been higher.

The *in vivo* digestibility values for the three precooked samples(squid, shrimp and pollock) were all very close, with values above 90% as shown in

**Table 4. Apparent *in vivo* digestibility of precooked seafoods diets**

Diet	Total consumed feed (g/10 rats)	Collected feces		Total excreted protein (g)	<i>In vivo</i> digestibility (%)
		Wt. (g/10 rats)	Protein(%)		
Casein (ANRC)	1342.3	51.8	21.90	11.3	91.9
Squid	1512.2	59.5	21.20	12.6	92.2
Oyster	1024.0	72.5	20.94	15.2	85.5
Shrimp	1594.5	60.7	20.53	12.5	92.3
Pollock	1456.1	53.0	21.34	11.3	92.2
Laver	1048.7	128.5	17.36	22.3	79.0

Table 4. In contrast, the *in vivo* digestibilities for oyster showed 85.5% and 79.0% for the laver. Squid and shrimp *in vivo* digestibilities were 6 to 7% lower than the results obtained by Iwaya and Yamaguchi(1979).<sup>18)</sup> True protein digestibility of protein food always higher than apparent digestibility<sup>36,37)</sup> since true protein digestibility account the metabolic nitrogen which is not of dietary origin. Oyster and laver both possessed lower *in vivo* digestibilities than did squid, pollock and shrimp. It has been known that oyster are a good source of protein, exhibiting high protein digestibility.<sup>25)</sup> But in this study, the *in vivo* digestibility of freeze dried whole oyster(stored for 3 months at 5°C) used in rat assay was found to be inferior to casein and the other seafood diets. This can be accounted for noting the oxidized fat formed during processing and the influence of storage on the rat's digestibility. The rancid odor of oyster diets caused a sharp decrease in appetite and diarrhea in the rat, during *in vivo* digestibility assay(see Table 4). Oxidized unsaturated lipids in oyster bind to proteins and form insoluble lipid-protein complexes.<sup>38)</sup> The proteins of seaweed are poorly utilized when the intact cells are fed to animals,

although the isolated algae or seaweed protein can easily be digested.<sup>31,39)</sup> The results based on previous reports<sup>40,41)</sup>, indicate that disrupting the cell wall of laver in the mill caused an increase of *in vivo* digestibility, giving it a value which is comparable to that of algal protein.<sup>32,42-44)</sup>

In order to assess the overall protein nutritional quality, ie. C-PER, DC-PER and predicted digestibility for the precooked seafoods, the amino acid composition of seafoods were determined and reported in Table 5. If the amino acid profile of the squid sample was compared with the previous results<sup>9,14,45)</sup>, it was found that there were slight variations in methionine and tryptophan contents which could probably due to 1) the analysis procedure used(previous, tryptophan results were obtained by Spies method<sup>46)</sup> and 2) the decrease in methionine being a result of heat processing.<sup>47,48)</sup> The limiting essential amino acid in oysters was tryptophan and its chemical score was 52.8, which was comparable to 86(Trp) from Matsuno(1973).<sup>28)</sup> Levels of all amino acids(especially in nonessential amino acid) in precooked oyster were higher than that observed in raw oyster.<sup>14)</sup> High levels of the nonessential amino acids in precooked oyster could

Table 5 Amino acid profiles (g a. a. /16 g N.) precooked seafoods

Amino acid	ANRC Casein	Squid	Oyster	Shrimp	Pollock	Laver
Asp	7.12	11.19	13.12	10.70	10.75	9.77
Thr	4.08	4.83	6.04	4.16	4.70	5.79
Ser	5.27	4.80	6.24	4.37	5.12	5.52
Glu	22.72	16.43	17.89	16.73	15.59	11.40
Pro	11.00	3.71	5.90	3.27	3.29	4.39
Gly	1.83	4.73	6.78	4.40	4.60	6.84
Ala	3.08	5.48	6.71	5.63	5.85	12.12
Val	6.60	4.22	6.53	4.57	5.04	5.89
Met	2.84	3.04	3.75	3.93	3.46	2.62
Ile	5.25	4.59	5.10	4.98	4.32	3.57
Leu	9.66	8.67	9.03	8.43	8.37	7.48
Tyr	5.66	3.81	4.52	3.71	3.67	3.39
Phe	5.21	4.38	4.99	5.47	4.21	4.20
Lys	8.23	8.84	5.47	9.24	9.98	4.43
His	2.90	2.20	1.41	2.13	2.12	1.34
Amm	1.94	1.27	1.10	1.22	1.36	1.75
Arg	3.87	8.05	4.78	8.39	7.30	6.24
Cys	0.58	1.31	1.55	1.21	1.40	1.86
Trp	1.03	0.74	1.04	0.93	0.93	1.00

be accounted for by noting the concentration and structural changes resulting from denaturation, releasing ammonia and coagulation of proteins as affected by cooking. The decrease of lysine and cystine content after cooking was similar to the results mentioned by Miller et al. (1965).<sup>48)</sup> The most limiting essential amino acids in oyster sample were histidine and tryptophan and oyster's chemical score was 54.1 for rats, and 74.3 for human. Those results were comparable to the data on chemical score 63(Trp).<sup>28)</sup> It was also interesting to note that all essential amino acids(except cystine and tryptophan) of peeled and deveined shrimp were higher than the data in previous reports<sup>14, 15, 45)</sup>, while nonessential amino acids were lower than those of above references, which was expected since it is known that these amino acids are easily degraded by heat. The limiting amino acid was also tryptophan and its chemical score was 66.4. The essential amino acid composition of precooked pollock fillet was also similar with that of pollock FPC.<sup>17, 18)</sup> Slight differences were observed as compared Mihara et al. (1977). This difference could probably be due to sample preparation. The 9.98 (g/16 gram N.) of lysine found in pollock fillet was the highest lysine content among the samples used in the present study and it was superior to casein. The limiting amino acid was tryptophan, and its chemical score was 66.4. The proximate composition and amino acid profiles of seaweed were dependent on the origin of sample, the collection time, and species.<sup>20)</sup> Laver was showed excellent EAA profiles when compared with the previous data on EAA composition of marine algae.<sup>39, 44, 49, 50, 51)</sup> The lysine content of the laver was low, typical of many plant proteins. Lysine was the limiting amino acid in laver and it showed 69.6% requirement vs % for that reported by Matsuno (1973).<sup>28)</sup>

*In vivo*, *in vitro* and digestibility estimated solely from the amino acid profile, are shown in Table 6. The squid, shrimp and pollock samples had comparable *in vivo* digestibility values, approximately equal to that of the reference protein casein(about

**Table 6. Apparent *in vivo* and *in vitro* digestibility and predicted digestibility of precooked seafood diets**

Sample	<i>In vivo</i> <sup>a</sup> digestibility (%)	<i>In vitro</i> <sup>b</sup> digestibility (%)	Predicted <sup>b, c</sup> digestibility (%)
ANRC casein	91.9	90.3	87.2
Squid	92.2	88.5	90.5
Oyster	85.5	80.2	92.1
Shrimp	92.3	88.1	91.7
Pollock	92.2	86.2	93.1
Laver	79.0	81.2	85.3

<sup>a</sup> Pooled mean from 10 rats per treatment

<sup>b</sup> Samples were stored for 3 months at 4°C after precooking and freeze drying

<sup>c</sup> Estimated solely from the amino acid profile

92%). In contrast, the oyster and laver were noted to have *in vivo* digestibilities 6~11% lower than that of casein. The same pattern was revealed in the *in vitro* assay, which showed samples with high *in vivo* digestibilities also possessed high *in vitro* digestibilities and it showed that the *in vitro* assay was underestimating digestibility as compared to *in vivo* assay. Oyster showed the lowest *in vitro* digestibility(80.2%) compared to *in vivo* digestibility of 85.5%. All the animal protein samples had *in vitro* digestibilities which were lower than their *in vivo* digestibilities. This small but consistent discrepancy between *in vitro* and *in vivo* digestibility was discussed by Satterlee et al. (1979).<sup>52)</sup> In case of laver, it is noted that *in vitro* digestibility was higher than the *in vivo* digestibility. The differences noted the *in vitro* and *in vivo* values could be due to several reasons. First, the *in vitro* digestibility assay may be more sensitive to small chemical changes in protein structure than the rat. A small increase in peptide bond susceptibility to enzymatic attack may result in a large increase *in vitro* digestibility, while being relatively ignored by the rat.<sup>53)</sup> Secondary, it was difficult to adjust the pH of laver sample due to the high viscosity when in solution. This may have caused the pH meter electrode to drift, resulting in possible errors in the pH values measured. Since the calculation of the *in vitro* assay is based on the pH drop from 8.0, any failure to properly adjust the initial

samples would have greatly affected the final results. In case of predicted protein digestibility, samples with high *in vivo* and *in vitro* digestibility also showed high predicted digestibility. However for oyster, the predicted digestibility was 92.1% while *in vitro* and *in vivo* digestibility was 80.2% and 85.5%, respectively.

As shown in Table 7, the values of C-PER were very close to the rat-PER values, except for the squid sample. However, C-PER values were slightly lower than the rat-PER, but in case of shrimp and pollock it was in contrast with previous reports.<sup>54,55)</sup> The greatest difference was found between rat-PER (2.9) and C-PER (1.9) or squid. The proteins of such samples were well digested *in vivo*, were somewhat resistant to enzymatic hydrolysis by the *in vitro* assay, unexplainable problems in amino acid profiles could also be the cause of discrepancy. In case of oyster, the C-PER value was slightly higher than its rat-PER, indicating that the influence of enzyme indigestible substrates in the oyster sample on rat's *in vivo* digestibility is greater than that on *in vitro* digestibility by enzyme hydrolysis. The higher value of C-PER for laver demonstrates that the *in vitro* digestibility of laver was overestimated as mentioned above. The correlation coefficient between C-PER and rat-PER was 0.5021. The relatively poor correlation could be attributed to the low C-PER value of squid that we believe was an outlier which biased the data. If the datum for squid was eliminated from C-PER data comparisons, the C-PER correlated highly with rat-PER ( $r=0.9524$ ). In contrast to the bias of C-PER, DC-PER values of samples were closer to the rat-PER ( $r=0.9603$ ) but the difference between C-PER value

and rat-PER was slightly greater than the difference between DC-PER and rat-PER. According to the results in Table 7, it would appear that the seafood samples which possess a high protein digestibility, such as squid, shrimp or pollock, need DC-PER procedure rather than C-PER procedure to predict the protein quality as mentioned by Satterlee et al. (1981).<sup>52)</sup>

## Summary

A study was undertaken to evaluate the nutritional quality of protein from precooked seafoods. Procedures for evaluation included protein efficiency ratio(PER) using the rat, computed PER(C-PER) and discriminant computed PER(DC-PER) techniques. These procedures involve the determination of *in vitro* digestibility and amino acid composition of the sample prior to computation of C-PER and DC-PER. The values of C-PER for squid, shrimp and pollock were slightly lower than the rat-PER, while C-PER value in laver was higher. For the oyster, the C-PER value was very close to the PER value obtained from the rat assay. The difference between DC-PER value and rat-PER or NPR was slightly lower than that between C-PER and rat-PER except oyster and laver. Seafood samples which possess a high *in vitro* protein digestibility may need the DC-PER procedure rather than C-PER procedure. The C-PER procedure could offer more advantages in predicting the protein quality of seafood samples than the DC-PER procedure which showed poor *in vitro* digestibility.

## References

1. Ryu, H.S. and Lee, K.H.: *J. Korean Soc. Food Nutr.*, **14**(1), 1(1985)
2. AOAC: "Official Methods of Analysis", 12th ed., Association of Official Analytical Chemists, Washington D. C. (1975)
3. Dunlap, C.J., Guadagni, D.E., Miers, J.C., and Wagner, J.R.: *Food Prod. Devel.*, **8**, 88 (1974)
4. Bender, A.E. and Doell, B.H.: *Brit. J. Nutr.*,

**Table 7. Comparison of C-PER, DC-PER, rat PER, and NPR of precooked seafoods**

Sample	NPR	Rat-PER	C-PER	DC-PER
ANRC casein	4.8	2.5	2.5	2.5
Squid	5.8	2.9	1.9	2.9
Oyster	4.2	1.9	2.0	2.1
Shrimp	5.6	2.9	2.6	2.7
Pollock	5.8	3.0	2.6	2.7
Laver	4.0	1.7	2.1	2.1



- 11, 14(1957)
5. Osborne, T.B., Mendel, L.B., and Ferry, E.L.: *J. Biol. Chem.*, **37**, 222(1919)
6. AOAC: *J. AOAC*, **65**, 496(1982)
7. Hugli, T.E. and Moor, S.: *J. Bio. Chem.*, **247**, 2828(1972)
8. Moor, S.: *J. Bio. Chem.*, **238**, 238(1963)
9. Lee, C.M., Toledo, R.T., Nakayama, T.O.M., and Chichester, C.O.: *J. Food Sci.*, **39**, 735 (1974)
10. Suyama, M., Konosu, S., Hamabe, M., and Okuda, U.: In "Utilization of Squid" (in Japanese), Kosesha Kosekaku, Tokyo, 209(1981)
11. Borgstrom, G.: In "Fish as Food", Vol. II, Academic Press, New York, 121(1965)
12. Hatanaka, M.: *Bull. Jap. Soc. Sci. Fish.*, **9**, 21 (1940)
13. Gordon, D.T. and Roberts, G.L.: *J. Agric. Food Chem.*, **25**, 1262(1977)
14. Mihara, T., Suzuki, I., and Iwami, D.: In "Handbook of Food Analysis" (in Japanese), Kenpakusha, Tokyo, 760(1977)
15. Shrinivas, H., Vakil, U.K., and Shreenivasan, A.: *J. Food Sci.*, **39**, 807(1974)
16. Toma, R.B. and James, W.H.: *J. Agric. Food Chem.*, **23**, 1168(1975)
17. Lee, E.H. and Kim, S.K.: *Bull. of Korean Fish. Soc.*, **12**, 103(1979)
18. Iwaya, M. and Yamaguchi, M.: *Jap. J. Nutr.*, **37**, 247(1979)
19. Power, H.E.: *J. Fish. Res. Board Canada*, **21**, 1489(1964)
20. Ogino, C.: *J. Tokyo Univ. Fish.*, **41**, 110 (1955)
21. Ryu, H.S. and Lee, K.H.: *Bull. of Korean Fish. Soc.*, **10**, 151(1977)
22. Yoshimura, K. and Sakan, N.: *Bull. Fac. Fish. Hokkaido Univ.*, **3**, 205(1954)
23. Velera, G., Pujol, A., and Moreiras, O.: In "Fish in Nutrition", Fishing News(Books) Ltd., London, 259(1961)
24. Matsuno, M. and Iwaya, M.: *Jap. J. Nutr.*, **29**, 250(1971)
25. Lanham, W.B. Jr. and Lemon, J.M.: *Food Research*, **3**, 549(1938)
26. Schwarz, M.G. and Watts, B.M.: *Food Research*, **22**, 76(1957)
27. Johnes, D.E.: *Am. J. Pub. Health*, **14**, 177 (1926)
28. Matsuno, M.: *Jap. J. Nutr.*, **31**, 262(1973)
29. Sidhu, G.S., Montgomery, W.A., Holloway, G.L., Johnson, A.R., and Walker, D.M.: *J. Sci. Food Agric.*, **21**, 293(1970)
30. Morrison, A.B., Sabry, Z.I., and Middleton, E.J.: *J. Nutr.*, **77**, 97(1962)
31. Hedenskog, G. and Enebo, L.: *Biotech. and Bioeng.*, **11**, 37(1969)
32. Lubitz, J.A.: *J. Food Sci.*, **28**, 229(1963)
33. Henry, K.M.: *Brit. J. Nutr.*, **19**, 125(1965)
34. Rao, S.V., Daniel, V.A., Joseph, A.A., Sankaran, A.N., and Swaminathan, M.: *J. Nutr. and Diet.*, **1**, 103(1964)
35. McLaughlin, J.M.: In "Soy Protein and Human Nutrition", Academic Press, New York, 281 (1979)
36. Bodwell, C.E., Satterlee, L.D., and Hackler, L.R.: *Am. J. Clin. Nutr.*, **33**, 677(1980)
37. Hopkins, D.T.: In "Protein Quality in Humans", AVI Pub. Co. Inc., Westport, 169(1981)
38. Roubal, W.T. and Tappel, A.C.: *Arch. Biochem. Biophys.*, **113**, 5(1966)
39. Woo, S.I., Ryu, H.S., and Lee, K.H.: *Bull of Korean Fish. Soc.*, **12**, 225(1979)
40. Kang, M.H.: *Research of Food Nutrition*(E-Hwa Univ.), **6**, 29(1976)
41. Larsen, B.A. and Hawkins, W.W.: *J. Sci. Food Agric.*, **12**, 735(1961)
42. Cook, B.B., Lau, E.W., and Bailey, B.M.: *J. Nutr.*, **81**, 23(1963)
43. Clement, G., Giddly, C., and Menzi, R.: *J. Sci. Food Agric.*, **18**, 497(1967)
44. Narashima, D.L.R., Venkataraman, G.S., Duggal, S.K., and Eggum, B.O.: *J. Sci. Food Agric.*, **33**, 456(1982)
45. Konosu, S., Katori, S., Ota, R., Eguchi, S. and Mori, T.: *Bull. Jap. Soc. Sci. Fish.*, **21** 1163(1956)
46. Spies, J.R. and Chamber, D.C.: *J. Biol. Chem.*

- 191, 1781(1951)
47. Sawant, P.C. and Magar, N.G. : *J. Sci. Food Agric.*, **12**, 347(1961)
48. Miller, E.L., Hartley, A.W., and Thomas, D.C. : *Brit. J. Nutr.*, **19**, 565(1965)
49. Smith, D.G. and Young, E.G. : *J. Biol. Chem.*, **217**, 845(1955)
50. Mazur, A. and Clarke, H.T. : *J. Biol. Chem.*, **123**, 729(1938)
51. Barta, A.E., Branen, A.L., and Leung, H.K. : *J. Food Sci.*, **43**, 1543(1981)
52. Satterlee, L.D., Kendrick, J.G., and Miller G.A. : *Food Tech.*, **31**, 78(1977)
53. Eicher, N. : MS thesis of Univ. of Nebraska Lincoln(1982)
54. Babji, A.S., Froning, G.W., and Satterlee L.D. : *J. Food Sci.*, **45**, 441(1980)
55. Jewell, D.K., Kendrick, J.G., and Satterlee L.D. : *Nutr. Reports Int.*, **21**, 25(1980)