

EVALUATION ON UTILIZATION OF SMALL MARINE FISH TO PRODUCE SURIMI USING DIFFERENT CRYOPROTECTIVE AGENTS TO INCREASE THE QUALITY OF SURIMI

Tri Winarni Agustini^{*1)}, YS. Darmanto and Danar Puspita Kurnia Putri

Fisheries Department, Faculty of Fisheries and Marine Science – Diponegoro University
Semarang 50275, Indonesia

Received : March, 17, 2008 Accepted : May, 26, 2008

ABSTRACT

Quality of surimi (minced meat of fish) usually decrease due to denaturation of protein. Addition of cryoprotective agent such as reducing sugar as anti denaturation of protein is very important during storage of frozen surimi. Recently, development of human lifestyle require healthy food such as utilization of stevia sugar (*Stevia rebaudiana*) which has low calorie that can be used to replace sorbitol and sucrose during processing of surimi. The purposes of this research are to observe the effect of different cryoprotective agents before and during storage as well as the effect of storage on quality of frozen surimi fish based on pH value, water holding capacity (WHC), gel strength and organoleptic value. Materials used was surimi made from kurisi (*Nemipterus sp.*), stevia sugar, sorbitol and sucrose. Research method used was experimental laboratory with research design of Completely Randomized Design with split plot in time. The main plot was difference of cryoprotectant (stevia sugar 0.6%; sorbitol 4%; sucrose 4%). The product was analysed every 15 days starting from 0 to 45 days storage at -10°C . The results showed that different cryoprotectants agent gave no significant effect before storage treatment proceed. In addition during storage, the different of cryoprotectant gave significant effect to WHC and gel strength (SSS=1356,416; SS=1458,525; S=1511,307 g.cm) but not for pH. The organoleptic value for appearance on 15 days storage was SSS=7; SS=7; S=6,56 and Folding test showed SSS=7; SS=7.78; S=7,89)

Key words: frozen surimi, cryoprotective agent, sucrosa, sorbitol, stevia sugar, quality

*Correspondece : Phone : +62-24-8310965; Fax : +62-24-7460039; email: tagustini@yahoo.com

INTRODUCTION

Surimi is one of fish processing products which is considered as *intermediate product*. It has high potential utility for product development of fisheries resources. Utilisation of surimi as raw material increase every year such as in Japan account for 17.658 million tons/year, Amerika 2.320 million tons/ year and Korea 1.082 million tons/year, therefore it has good market

penetration (BPPMHP, 2005). Development of surimi industry in Indonesia is fairly good even though some of them are still under supervision. Surimi can be made from any kind of fish especially it is recommended to utilise non economic fishes. Surimi can then be processed into several development products such as fish meat ball, fish sousage, fish nugget etc.

Deterioration of frozen products generally due to protein denaturation, dehydration, and lipid oxidation. Protein denaturation often occurs in surimi during frozen storage. Raw material used for surimi production is fresh white flesh fishes such as gulamah, kurisi, lizard fish, Cunang, cat fish, Pisang-pisang, Gabus and ray fish. Kurisi fish is one of white flesh fish that is often used for surimi production because it has high content of myofibrillar protein which results in good gel strength.

Apart from raw material, cryoprotectant also has significant effect to the quality of surimi. *Cryoprotectant* is a substance that act as anti denaturing agent during frozen storage. Sugar as cryoprotective agent can increase water shear surface to protect loss of protein molecule. Addition of cryoprotectant can improve quality and water holding capacity of surimi.

Development in food technology introducing stevia sugar (*Stevia rebaudiana*) as material which has sweet taste (200-300 times to sucrose) and is safely consumed and less calorie. Utilization of stevia sugar is rare in fisheries industry.

The effect of stevia sugar addition to the quality of surimi will be evaluated for pH value, WHC, gel strength, final organoleptic value of surimi. All parameters are analyzed during storage of frozen surimi with interval of 0, 15, 30, and 45 days.

The objectives:

1. To observe the effect of using different cryoprotective agents (stevia, sorbitol and sucrose) to the quality of surimi resulted based on organoleptic value, gel properties and chemical properties before storage.
2. To observe the effect of using different cryoprotective agents (stevia, sorbitol and sucrose) to the quality of surimi resulted based on organoleptic value, gel properties and chemical properties during frozen storage.

3. To observe the effect of storage time to the quality of frozen surimi of kurisi fish (*Nemipterus sp.*) from the view point of pH value, WHC and gel strength.

MATERIAL AND METHODS

Material used in this experiment are kurisi fish (*Nemipterus Sp*) as raw material, stevia sugar, sorbitol, sucrose, STPP and ice flake.

Preliminary Research

Preliminary research was conducted to get the best concentration of stevia sugar as cryoprotectant in processing of frozen surimi. Concentration used were 0%, 0,2%, 0,4%, 0,6%, 0,8%, 1%.

Test for *gel strength* of surimi was carried out after freezing of surimi (after 24 hours). Organoleptic test of the product was done using standard of SNI 01-2345-1991.

Main Research

This research used 3 type of cryoprotectants namely stevia sugar (S) using the best concentration, sorbitol (SS) with 4% concentration and sucrose (SSS) with 4% concentration. Sucrose and sorbitol are commonly used for surimi production. In this research, effectivity of stevia sugar will be compared to that of sucrose and sorbitol in surimi production.

Analysis of surimi product will cover:

- pH analysis (Suzuki, 1981)
- *Water Holding Capacity* (WHC) analysis (Hamm, 1975)
- *Gel strength* analysis (Suzuki, 1981)
- Organoleptic analysis for fresh fish (SNI-01-2345-1991)
- Organoleptic analysis for frozen surimi (SNI-01-2694-1992)

Experimental methods used was experimental laboratoris. This experiment based on observation which was planned to

get new fact or to strengthened and disagree the fact previously observed (Srigandono, 1981). Experimental design used was Completely Randomized Design with split plot in time, where storage time (0, 15, 30 and 40 days) as main plot and different cryoprotectants (Stevia sugar, sucrose and sorbitol) as sub plot with each treatment was conducted for 3 replication. Hypothesis was analysed using Honestly Significant Different test.

RESULTS AND DISCUSSION

Analysis of Raw Material

Raw material of Kurisi fish was taken from Semarang coastal area for approximately 15 kg with average length of 21 cm ± 2 cm and average weight of 80 gr ± 2 gram. The raw fish was then analysed for organoleptic using SNI 01-2729-1992 by 10 panelist (Appendix 2). The result for organoleptic showed average of 7,75 ($7,50 \leq \mu \leq 7,99$) for all spesification includes eyes, gill, slime, flesh and belly, odor, texture. Based on the

results it can be concluded that raw fish used in this experiment was considerably good enough because still acceptable with value above the minimum standard for fresh fish i.e 7. (Indonesian National Standard, 1992. Fish which is used for surimi production should be fresh, no phisical damage, prime quality because quality of actin and myosin in protein tissue of fresh fish still high as weel is its water holding capacity. (Perangin-angin, 1999).

Analysis of Frozen surimi of Kurisi fish

Analysis of frozen surimi start at 0 day storage and continue for every 15 days interval. Analysis was conducted before and after frozen storage for its pH value, WHC, gel strength and organoleptic of frozen surimi.

pH of frozen Surimi

Initial pH of surimi for each treatment during storage showed no significant different ranging from 6.93 - 7 at 0 day storage and 7.47 - 7.67 at 45 days storage. (**Table 1**).

Table 1. The pH of Frozen Surimi of Kurisi fish with addition of different cryoprotectant

Storage time (days)	<i>Cryptotectant</i>		
	SSS	SS	S
0	6.93±0.058	6.97±0.058	7±0.100
15	7.27±0.058	7.27±0.058	7.27±0.115
30	7.43±0.115	7.33±0.208	7.37±0.058
45	7.67±0.058	7.57±0.0578	7.47±0.058
Average	7.325±0.308	7.283±0.247	7.257±0.200

Note : Value is an average of 3 replication ± SD

SSS : Sucrose SS : Sorbitol S : Stevia

Based on analysis of variance (appendix 3), different cryoprotectant did not give significant effect ($F_{hit} < F_{table}$ 0,05) to pH of frozen surimi before storage, after storage and its interaction. In addition,

storage time gave significant effect ($F_{hit} > F_{table}$ 0,01) to pH of frozen surimi.

Based on HSD test it was concluded that every treatment did not give significant effect on pH of frozen surimi, except for treatment of sucrose to stevia. Both gave

significant effect to pH of frozen surimi. Storage time showed that starting from 0 day storage to 45 days storage, each

treatment give significant effect to pH of frozen surimi of kurisi fish.

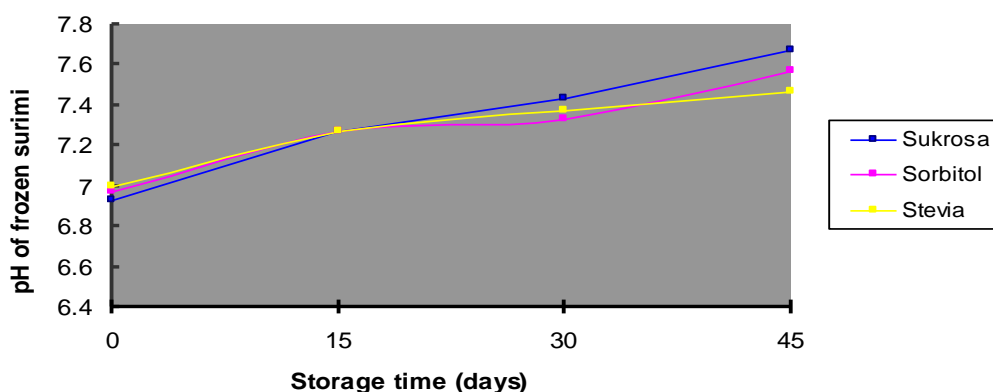


Fig 1. The effect of different *Cryoprotectant* (Sucrose, Sorbitol, Stevia) to pH of frozen surimi during storage

Based on the results, pH value of surimi before storage ranging from 6.94 – 7. This results was similar to pH resulted from Hanggiani (2004) with pH 6.9 – 7. From this result, different cryoprotective agent did not affect pH of surimi at 0 day storage. However, during storage time, the pH of surimi showed increase gradually for each treatment. Surimi added by Stevia sugar had lower pH compared to Sorbitol and Sucrose after storage for 45 days. pH fluctuation of surimi during storage ranging from 7.45 – 7.67 which considered normal. This is because myofibril protein not stable only when pH less than 6.5 due to loss in ATPase enzym activity. (Tanaka, 2001). At pH \pm 6, gel properties resulted will be more compact and homogen and when pH > 8, gel resulted will be relatively low and not homogen. Increased pH of frozen surimi during storage at temperature of $-10^{\circ}\text{C} \pm 3^{\circ}\text{C}$ in this research related to protein degradation process which resulted in amin and amoniak.

Water Holding Capacity of frozen Surimi

Average WHC for each treatment ranging from 26.530% - 33.103% during storage of surimi. The highest WHC of surimi obtained on surimi treated by stevia and the lowest was treated by sorbitol. Based on analysis of variance, it was found that different cryoprotective agent, storage time and interaction between them give significant effect ($F_{hitung} \geq F_{tabel}$ 0.01) on WHC of frozen surimi.

According to HSD test, different cryoprotective agent give very significant effect between Stevia sugar and either sucrose treatment and sorbitol treatment. But there was no significant effect between sucrose treatment and sorbitol on WHC of frozen surimi. Decreased WHC of frozen surimi during storage was due to denaturation protein occurred in the product which give influence on decreasing hydrophilic properties of protein and

resulted on reducing myofibrillar protein capacity to form hydrogen bonding with water (Sen *et al.* 1981). Decreased on WHC of frozen surimi during storage can be seen on Fig.3.

When pH of surimi tend to reach alcali condition, hydrogen bonding will gradually decrease and more water release

from surimi. If this surimi was then heated, the texture resulted is not tough and soft. Addition of sugar will then improve water holding capacity of myofibrillar protein because sugar can increase surface stress of protein molecule so that water can retain in the tissue.

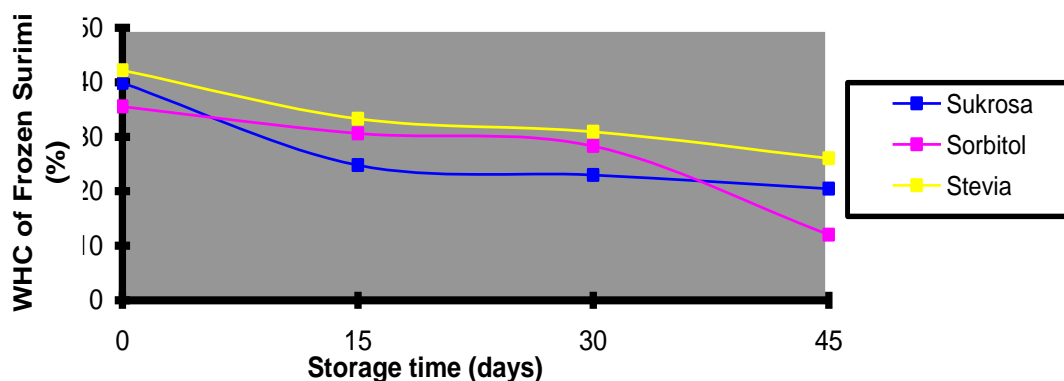


Fig 2. The effect of different Cryoprotective agent (Sucrose, Sorbitol and Stevia) to WHC of Frozen surimi during storage

Sugar as cryoprotective agent is supported by the presence of sodium tripoliphosphate that affect in retarding myofibril protein dissolve. Okada (1963) stated, adding of poliphosphate can increase texture and humidity due to increase of pH, ionic strength and protein interaction.

Stevia has proven can retain water in protein molecule better than sucrose and sorbitol. This occurred because stevia has more complex in molecular structure compared to sucrose and sorbitol which resulted in more protein molecule can form hydrogen bonding with water.

According to Lanier and McDonald (1991), mechanism of *cryoprotectant* was discovered can stabilised protein when react with water surrounding. This imply on protein can be protect from dehydration during freezing and frozen storage. This phenomena supported by Gopakumar (1997), basically can improve surface stress of water and the number of water bonded as

well as protect the product from drip loss so that protein molecule will be more stable.

Gel Strength of Frozen Surimi

To analyse physical properties of surimi can be done by measuring gel strength. Before measuring gel strength, the surimi should be converted to kamaboko product. Physical properties of gel surimi can be analysed only when the kamaboko product is heated. Heating temperature is very important in setting gel of kamaboko. Good kamaboko give high gel strength.

The results showed that average gel strength of treatment ranging from 1356.416 – 1511.307 g.cm. Based on analysis of variance, different cryoprotectant has no significant effect to gell strength before storage. However, different cryoprotectant and storage time give significant effect to gel strength during storage ($F_{hitung} \geq F_{tabel}$ 0.01). Interaction between those two factors

also give significant effect to gel strength of surimi.

Based on HSD test, different cryoprotectant give significant effect to gel

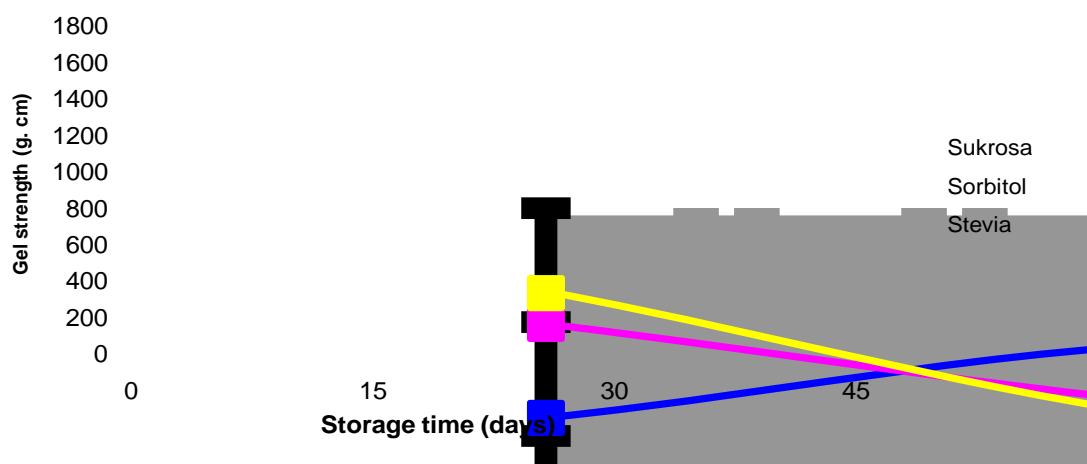


Fig 3. The effect of different cryoprotectant (Sucrose, sorbitol, stevia) on Gel strength of surimi during storage

Different sugar cause different on gel strength fo surimi resulted. Even though stevia, sorbitol and sucrose are belong to glicoside but they have different molecular structure. Sugar act to bind water and it can prevent gel elasticity of surimi. The better the activity of sugar, the better its capability to form gel of protein. Stevia has molecular structure of $C_{38}H_{60}O_{18}$, sorbitol $C_8H_{14}O_6$ and sucrose $C_{12}H_{22}O_{11}$. Different in molecular structure will affect on water holding capacity in protein and water bonding. Hydrogen bonding and hydrophobic influence on initial gel formation (gel setting) where this bonding help in prevention surface stress on protein tissue (Okada, 1963; Niwa dan Miyaka, 1971). Therefore WHC is an important factor determining in gel formation of kamaboko. Higher WHC of protein, better gel formation of kamaboko.

Gel strength value resulted from the research are categorized as high gel strength until the end of storage (45 days). This can be explained that raw material used in this experiment was kurisi fish which is white

strength of surimi during frozen storage. **Fig. 4** showed decreased of gel strength during storage fo frozen surimi.

flesh fish and has high protein content of 12%. This condition can result in good gel strength of product because myofibrillar protein especially actomyosin play an important role on gelling formation.

In addition geeling formation also affect by heating method applied on the surimi based product. Heating method by applying 2 steps i.e first step heating at $< 50^{\circ}C$ (setting temperature) to form "sol state" becoming "suwari state". Second step is heating at $> 80^{\circ}C$ to cook the gel formed to become strong fish paste. (Sarifah, (1996) in Kohar (2004). Suzuki (1981) stated further that slow heating in the initial state at temperature $> 50^{\circ}C$ will result in lower elasticity of actomyosin.

Organoleptic Analysis of frozen surimi.

Folding Test

According to Lanier (1992), folding test is suitable assessment for gel product to differentiate between products with good and poor quality of gel. However this

method is not sensitif to differentiate between product with good quality (A) and excelent (AA) because almost shos the same value. The folding test data for kamaboko produced from surimi with different treatment of cryoprotectant is shown in (Fig 4).

After 45 days storage, the data obtained for folding test of the sample ranging from 6.78 – 6.89 which are closely to grade A (Value of 7). The similar results also found in surimi produced from the same fish (Kohar, 2004).

From (Fig. 4), it can be explained that decrease in folding test value during storage was due to protein denaturation especially myofibrilar protein that affect the formation of gel. In addition water content of surimi has also important influence and can be used as indicator of folding test value. Water content of surimi resulted from this research after 45 days storage ranging form 80 – 82%. According to Suzuki (1981), water content of mu-en surimi of 81.5% result in folding test value of 5 (do not crack when it is folded into 4).

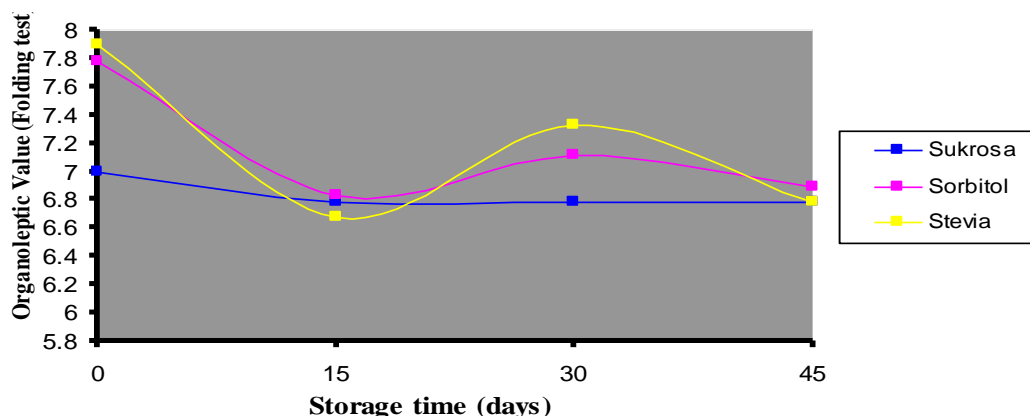


Fig 4. The effect of different cryoprotectant (Sucrose, sorbitol, stevia) on folding test of surimi during frozen storage

The folding test results showed that data obtained was not acurately measured since this method is categorised as subjective method. Lanier (1992) suggested that the folding test can only be applied and compatible to differentiate between product with good gel quality and poor gel quality but it does not sensitive to differentiate the product between good gel quality (A) and excellent gel quality (AA) because it gives similar results.

The folding test is also linier to the value of gel strength, where increase in gel strength is followed by increase in folding test value. Decrease in folding test during frozen storage of surimi is considered due to myofibrilar protein denaturation so that gel

formed becoming fragile. Sikorski and Pan (1994) stated that frozen storage can result in losses of functional properties of fish muscle protein for example gel formation of fish.

CONCLUSION

Based on the research on the effect of different cryoprotectant (sugar) in preventing quality of frozen surimi of kurisi fish during sotrage, it can be concluded that:

1. Different cryoprotectant (stevia, sorbitol, sucrose) has no significant effect on pH, WHC, gel strength and

- organoleptic value of frozen surimi of kurisi fish before storage
2. Different cryoprotectant (stevia, sorbitol and sucrose) has no significant effect on pH of frozen surimi during storage, but has very significant effect on WHC and significant effect on gel strength. Folding test of surimi showed significant effect on 15 days storage
 3. The storage time has very significant effect on pH, WHC, gel strength and organoleptic value of frozen surimi.

REFERENCES

- Arakawa, T. dan Thimaseft, S.N. 1982. Stabilization of Protein Structure by Sugars. *Biochemistry*.
- Borgstrom, G., 1965. *Fish as Food Vol IV Processing part 2*. Academic Press. United Kingdom.
- Buttkus, H. 1970. Accelerate Denaturation of Myosin in Frozen Solution. *J Food Sci.* 35 : 558-563
- BPPMHP. 2001. *Teknologi Pengolahan surimi dan Produk Fish Jelly*. Direktorat Jenderal Perikanan. Jakarta.
- BPPMHP. 2005. *Teknologi Pengolahan Surimi dan Produk Fish Jelly*. Balai Pengujian dan Pengawasan Mutu Hasil Perikanan (BPPMHP). Jakarta.
- BPPMHP. 2007. *Laporan Tahunan Balai Pengujian Mutu Hasil Perikanan Jawa Tengah Edisi 2007*. BPPMHP. Semarang.
- Coulter, T.P. 1990. *Food The Chemistry of Its Component, Second Edition*. The Royal Society of Chemistry. London.
- Chen J.C.P dan C.C. Chou., 1993. *Cane Sugar Hand Book*. John Wiley And Sons Inc. Canada.
- Darmawan, A. 2005. Sorbitol Pemanis untuk Penderita Diabetes. [http:// www. SuaraMerdeka. com](http://www.SuaraMerdeka.com). Cybernews 28 Februari 2005.
- Department Of Agriculture Philippines. 1995. *Production Of Surimi-Shrimp Value Added Products*. Bureau Of Fisheries And Departemen Of Agriculture Philippines and Aquatic Resources . Philippines.
- Direktorat Jenderal Perikanan. 1990. *Buku Pedoman Pengenalan Sumber Daya Perikanan Laut (Jenis-Jenis Ikan ekonomis Penting)*. Dirjen Perikanan, Departemen Perikanan. Jakarta.
- Direktori Pengusaha Sektor Kelautan dan Perikanan. Direktorat Jenderal Peningkatan Kapasitas Kelembagaan dan Pemasaran Departemen Kelautan dan Perikanan Edisi 2004. Dirjen Perikanan. Jakarta.
- Hanggiani, A. 2004. Pengaruh Lama Penyimpanan Surimi Beku dan Penambahan tepung Tapioka Terhadap Mutu Produk Kamaboko Ikan Kurisi (*Nemipterus sp.*). Skripsi (S1). Universitas Diponegoro. Semarang.
- European Commission. 1999. *Opinion of Steviosida as Sweetener*. Direktorat General European Commission of Food. UE.
- Fardiaz, D. 1985. *Kamaboko, Produk Olahan Ikan yang Berpotensi untuk Dikembangkan*. Media Teknologi Pangan, Volume I. Bogor.
- Gomez, K.A and A.A. Gomez. 1995. *Prosedur Statistik untuk Penelitian Pertanian*. Penerbit Universitas Indonesia. Jakarta.

- Gopakumar, K. 1997. Tropical Fishery Product. Science Publishes Inc. United Kingdom.
- Hardman, T.M., 1989. Protein Water Interaction dalam Water and food Quality. Ilyas, S. 1993. Teknologi Refrigerasi Hasil Perikanan Jilid II, Teknik Pendinginan Ikan. CV Paripurna. Jakarta.
- Hamm, R. 1975. Water Holding Capacity of Meat, in Meat. Butterworths. London.
- Hadiwiyoto, S. 1993. Teknologi Pengolahan Hasil Perikanan Jilid 1, Teknik Pendinginan Ikan. CV Paripurna. Jakarta.
- Honikel, K. O. 1987. How to Measur the Water Holding Capacity of Meat, Recommendation of Standarized Methods. M. Nijhoff Publisher. Netherlands.
- Instituto Agronomico. 2000. Journal of Stevia. IMECC Cx Press. Brazil.
- Kohar, K.P. 2004. Pengaruh Penambahan Tepung Tapioka terhadap Mutu Kamaboko dari Surimi Ikan Kurisi. Skripsi S1. Universitas Diponegoro. Semarang.
- Koswara, S. 2001. Surimi Suatu Alternatif Pengolahan Ikan. [http:// www. PagandanGizi. co. id](http://www.PagandanGizi.co.id). EBook Pangan.
- Lanier, T. and MacDonald, G.A. 1991. Carbohydrates as *Cryoprotectants* for Meats and Surimi. Food Science Depth. Nort Carolina USA.
- Lanier, T. C. 1992. Measuremnts of Surimi Composition and functional Properties in Surimi Process Technology. Marcel Decker Inc. New York.
- Lee, C.M. 1984. Surimi Process technology. Institute of food Technology. United State.
- Matsumoto, J. J. and Noguchi, S. F. 1991. Cryostabilization of Protein Surimi. In Surimi Teknologi, Editor Lanier, T. C. and C. M. Lee. Champman and Hall Publisher. New York-London.
- Okada. M. 1963. Ingredients On Gel Texture. Suzuhiro Kamaboki Kogyo Co. Japan.
- Okada, M. Chemistry of SURIMI Technology in Japan, dalam Surimi Teknologi. Editor lanier, T.C dan Lee, C.M. Marcel Decker Inc. New York
- Prayitno, E. 2003. Skripsi Kajian Proses Nugget dari Surimi Ikan Manyung (*Arius thalassinus*) dengan Bahan Tambahan Gelatin dari Kulit Ikan Tuna. Program Pasca Sarjana institute Pertanian Bogor. Bogor.
- Perangin-angin, Rosmawati. 1999. Teknologi Pengolahan Surimi. Balai Penelitian Perikanan Laut. Jakarta.
- Pomeranz, Y. 1991. Functional Properties of Food Components. Second Edition. Academic press. New York.
- Srigandono, B 1981. Rancangan Percobaan Experimental Design. Universitas Diponegoro. Semarang.
- Suprayatmi, S. 1996. Yang Manis Tidak Selalu Manis. [http:// www. PagandanGizi. co. id](http://www.PagandanGizi.co.id). Cybernews 18 Juli 1996.
- Steel, R.G.D. and J. H. Torrie. 1980. Principles and Procedures of Statistika a Biometrical Approach, 2nd Edition. McGraw Hill Kogasusha Ltd. Tokyo-Japan.
- Sarifah, S. 1996. Pemanfaatan Ikan Gurami (*Ospromemus gouramy Lac*) dalam

- Pembuatan Semi Produk Gel Ikan. Skripsi . Program Studi Teknologi hasil Perikanan, Fakultas Perikanan, Institut Pertanian Bogor. Bogor.
- Sen, L.C., Lee, S.H., Feeney, R.E and Whitaka, J.R. 1981. In Vitro Digesibility and Fuctional Properties of Chemically Modified Casein. *J. Agritech. Food. Chem* 27 : 811-818
- Sikorski, Z. E. and Pan, S. B. 1994. Preservation of Seafood Quality in F. Shaidi and J.R. Botta (eds.), *Seafoods Chemistry Processing Technology and Quality*. Blackie Academic and Professional London. United Kingdom.
- Simizu, Y., 1984. *Biochemical and Functional Properties of Material Fish*. Kyoto University. Kyoto, Japan.
- Sudarmadji, S 1992. *Bahan-Bahan Pemanis*. Agritech. Yokyakarta Soeparno. 1994. *Ilmu dan Teknologi Daging*. Gadjah Mada University Press. Yokyakarta.
- Sudarmadji, S. B. Haryono dan Suhardi. 1989. *Analisa Bahan Makanan dan Pertanian*. PAU Pangan dan Gizi Universitas Gajah Mada. Yokyakarta.
- Sumiono, B. 2000. *Pengkajian Perikanan Udang Penaeid di Laut Arafura*. Dalam *Jurnal Pasca Panen Perikanan* Vol 11. 2001. Balai Penelitian Perikanan Laut. Jakarta.
- Sun, C. T. and Wang, H. H. 1984. Cryoprotective Mechanism of Polyols and Monosodium Glutamate in Frozen Fish Mince. 3rd International congress on Enggland and Foods. Dublin.
- Suzuki, T.,1981. *Fish And Krill Protein Processing Technology*. Science Publisher LTD. London.
- Tanaka, M. 2001. *Surimi and Surimi Products*. Departemen of food Science and Technology. Jepang.
- Tanikawa, E., 1985. *Marine Product in Japan*. Kosisha Koseikaku Co, Ltd. Tokyo.
- Watanabe, T. 1990. *The Chemistry of Protein from Marine Animal in Science of Processing Marine Food product*. Japan International Agency. Japan.
- Winarno, F. G. 2004. *Kimia Pangan dan Gizi*. Gramedia Pustaka Utama. Jakarta.
- Wisper-Pedersen. (1971). *The Science of Meat and Meat Products* 2nd ed. W.H. Freeman and Co. Zayas, Joseph.F., 1997. *Functionality of Proteins in Food*. Springer-Verlag Berlin Heildeberg Publisher. Germany.
- Zayas, J.F. 1997. *Functionality of Proteins in Food*. Springer-Verlag Berlin Heidelberg. New York.