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A Study and Analysis of Materials, Methods and Research Methodology on Collection and Maintenance of Fish

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1. Introduction

Irrespective of the sex, live specimen of *Channa punctatus* measuring 15-18 cm in length and 30-35g of body weight were collected from local fish market. Fish were acclimatized to laboratory conditions for a period of two weeks. During acclimatization, fish were regularly fed on commercial fish food (Hikari circhild gold, KYORIN, Japan). Water was renewed after every 24h leaving no faecal matter, unconsumed food or dead fish if any.

2. Treatment of fish with toxic metals

Fish after acclimatization to laboratory conditions were divided into five groups. Each containing 10 fish. Group 1 containing 10 fishes were maintained in tap water to serve as control. Fish of Group 2 were treated with arsenic as arsenic trioxide (0.5ppm). Fish of Group 3 were treated with cadmium as cadmium chloride (0.5ppm). Fish of Group 4 were treated with chromium as chromium chloride (0.5ppm) and fish of group 5 were exposed to nickel as nickel chloride (0.5ppm) for total thirty-five days. These metallic compounds were procured from E. Merck, Mumbai.

3. Collection and staining of peripheral erythrocytes

After completion of exposure to respective metals/metalloids, and for desired duration fish were anesthetized by a mild blow on head. Peripheral blood samples were obtained from caudal vein of experimental fishes after 7 days, 14 days, 21 days, 28 days and 35 days of exposure to respective metals. Blood smears were prepared on clean slides, dried in air for 10 min & fixed in absolute methanol for 10 to 15 min. Fixed slides were stained in Giemsa for 45 min to 1h. Clear intact well stained cells were selected for calculating the MN frequency and other nuclear anomalies (Bolognesi and Hayashi, 2011) using light microscope (Nikon, Japan).

4. Criteria followed for micronuclei assay

Following criteria for micronuclei assay was followed during present study-:
1.Micronuclei must be less than one- third the diameter of main nucleus.
2.Must be same in focal plane.
3.Must have the same color, texture & representation as the normal nucleus.
4.Must be clearly separated from main nucleus.
5.Staining intensity must be similar to nucleus.

The average frequencies of all abnormalities (notched, blebbed, kidney shaped, segmented, lobed, bud shaped) were determined in each group. Average statistical mean ± SE was used for the description of the results.

Supply Chain Management (SCM) or supply chain management is a set of methods and approaches to improve integrity and efficiency between suppliers, manufacturers, warehouses, and stores so that

merchandise can be produced and distributed accurately both in terms of quantity, location and time (Levi, 2000; Aderinola, et.al. 2012; Aberoumand, 2014; Prakash, et.al. 2014; Baba, Sanchi & Manga, 2015).

SCM has actually been known for several years and is integrated with logistics. SCM confirms the interaction between marketing functions, production in a company. Utilizing the opportunity to improve services and reduce costs can be done through coordination and cooperation between the procurement of raw materials and distribution (Indrajit & Djokopranoto, 2002).

This is related to supply chain activities that are indirectly controlled by logistics activities. At present, it is undeniable that large and small companies must carry out logistics activities, both logistical within the company and outside the company. Channels of raw material supplies to the distribution of finished goods are in dire need of logistics (Siagian, 2005). These activities include traditional purchasing functions plus other important activities that relate between suppliers and distributors.

The component of SCM according to (Turban, 2004) consists of three main components, namely Upstream Supply Chain which includes the activities of a manufacturing company with its suppliers and the relationships between suppliers. Supplier relationships can be extended to several parts, starting with raw materials to finished goods. The main activity in the upstream supply chain is procurement. The second component is the Internal Supply Chain which includes all in house processes used in transforming inputs from suppliers into the manufacturing process. In the main internal supply chain activities carried out are the production, fabrication, and control of inventory. The last component is a downstream supply chain which includes all activities that involve the delivery of products to end customers. In the downstream supply chain, attention is directed to distribution, transportation warehousing, and after-sale service.

5. National Fish Logistics System

Regulation Number 5/KP/2014 explains that NFSL is a fish supply chain management system and fishery products as well as information ranging from procurement, storage, transportation to distribution, as a unity of policies to increase capacity and stabilize upstream-downstream fisheries production systems, control disparities and price stabilization, and to meet domestic consumption needs.

The purpose of NFSL is 1) Increasing the capacity and stabilization of the national fisheries production and marketing system; 2) Strengthening and expanding connectivity between upstream production centers, downstream production and marketing efficiently; 3) Improve the efficiency of fish supply chain management, materials, production equipment and information from upstream to downstream. Meanwhile NFSL component is same as the logistics system in general, which include purchasing process, storage, transportation, and distribution.

shows the operational NFSL scheme that has been set by the government through MMF. The role of the government is to provide new chain facilities and logistics infrastructure with the aim of supporting the smooth operation of NFSL. The intended value chain and logistics infrastructure facilities include fishing ports, container ports as sending ports, fishing fleet vessels, refrigerated storage units, marketing facilities, and others.

Many people view the sea as a land separator. Perspectives from the mainland's eyes have made us alienated and less utilizing the strength of the sea. This understanding gave rise to the idea of the sea toll. The sea toll is a concept of transporting marine logistics established by the Indonesian President, Joko Widodo. The sea toll lane program in question is to build sea transportation with ships or a logistics system, which serves non-stop from Sabang to Merauke, the main toll route can be seen

shows six sea toll lanes that connect between ports. The Ministry of Transportation in 2016 launched 6 scheduled sea transport routes that will connect large and small ports throughout the country to transport logistics and community needs.

The aim of the Sea Toll program is to move the economy wheel efficiently and evenly. One day there will be a big ship that will go back and forth in the Indonesian sea, so the logistics costs will be cheap. That is why sea toll is one of President Jokowi's priority programs to develop the maritime sector. One supporting factor is the need for a deep-sea port to provide a way for large vessels to cross the Sabang to Merauke route.

The Sea Toll Road is a track that stretches for 5,000 kilometers or one-eighth around the earth. The ideas on the Sea Toll program include 1) the establishment of a national maritime logistics system; 2) Creating equitable development; 3) Ensure economic connectivity between islands; 4) Streamline the distribution of goods, and 5) Suppress the price disparity. Aspects of the sea toll program include sea transportation aspects, port aspects, and shipping safety and security aspects.

6. Analytical Hierarchy Processes

The Analytical Hierarchy Processes (AHP) method was developed by Thomas L. Saaty, a mathematician. AHP methods help solve complex problems by structuring a hierarchy of criteria, interested parties, results and by attracting various considerations to develop weights or priorities. This method also combines the strengths of the feelings and logic involved in various problems, then synthesizes various diverse considerations into results that match our estimates intuitively as presented in the considerations that have been made (Saaty, 2005). AHP method is carried out with the following steps (Suryadi, 1998):

1. Define the problem and determine the desired solution. In this stage, we try to determine the problem that we will solve clearly, in detail and easily understood.
2. Make a hierarchical structure that starts with the main goal. After arranging the main goal as the top level, a hierarchy level will be arranged below, namely, the criteria that are suitable to consider or assess the alternatives we provide and determine these alternatives. Each criterion has a different intensity. The hierarchy is continued with sub-criteria (if possible).
3. Make pairwise comparison matrices. The matrix used is simple, has a strong position for a framework of consistency, obtains other information that may be needed with all possible comparisons and is able to analyze the sensitivity of the overall priority for changing considerations.
4. Perform and define paired comparisons. The results of the comparison of each element will be a number from 1 to 9 which shows a comparison of the importance of an element. The scale of the comparison of pairs and their meanings introduced by Saaty are as follows:

Intensity of Interest

- 1 = Both elements are equally important, two elements have the same effect;
- 3 = The one element is slightly more important than the other elements, Experience and assessment support a single element slightly compared to the other elements;
- 5 = The one element is slightly more important than the other elements, Experience and assessment support a single element slightly compared to the other elements;
- 7 = One element is more important than the other, Experience and judgment strongly support one element compared to the other;
- 9 = One element is clearly more important than the other elements, one element that is strongly supported and dominant is seen in practice;
- 2,4,6,8 = Values between two values of adjacent considerations, this value is given if there are two compromises between 2 choices;
- Reverse = If for activity i get one number compared to activity j, then j has the opposite value compared to i.

5. Calculate the eigenvalue and test its consistency, if it is not consistent then the data retrieval is repeated. Count steps 3, 4 and 5 for all levels of the hierarchy.
6. Calculate the eigenvector of each pairwise comparison matrix which is the weight of each element for prioritizing elements at the lowest hierarchy level until it reaches the goal.
7. Check hierarchy consistency. What is measured in AHP is the consistency ratio by looking at the consistency index. The expected consistency is near perfect so as to produce a decision that is almost valid. Although it is difficult to achieve perfect, the consistency ratio is expected to be less than or equal to 10%.

7. Preservation for Long Duration

7.1 Salting

There are many different kinds of salt, some being better than others for fish curing. However, in islands or in outlying places there is often no choice, and whatever is available in the way of salt has to be used, whether it is bought in a shop, prepared on the spot, or extracted from earth containing salt. A distinction must be made between the two chief techniques of salting: wet salting and dry salting (FAO, 2005).

7.2 Wet Salting

The principle is to keep the fish for a long time in brine. The equipment needed consists of a watertight container, which can be a tin, drum, canoe, barrel, etc. To make the brine, one takes four parts of clean water (sea or fresh water) and one part of salt. If the salt is coarse, it has to be ground or pounded first (Tys and Peters, 2009). It is then dissolved into the water by stirring with a piece of wood. To be good, the brine must float a fish. The next step depends on what kind of fish one wants to salt. It is best first to cut off the head, and gut and clean the fish, though small fish can also be salted whole. Large fish must be cut open, and it is preferable to take out the backbone. Fish with a heavy armour of scales must be scaled. In places where the flesh is thick, slashes must be made so that the salted brine can penetrate the flesh. Very large fish should be cut in thin fillets. After the fish has been prepared according to its size, it must be cleaned and put in the brine (FAO, 2008). A plank or matting is laid over it and weighted with rocks so that the fish is entirely covered with brine. This salted fish can be kept for a long time in a dark or at least a shady place (Leistner and Gould, 2002).

The remaining brine can be used three times, but water and salt must be added every time until a fish can again float on the liquid. In any case, fresh brine is always best.

7.3 Dry Salting

In this method the fish is salted but the juices, slime and brine are allowed to flow away. Dry salting can be done in an old canoe, or on mats, leaves, boxes, etc. In any case, the brine formed by the fish juices and the salt must be allowed to run away. For two parts of fish, one needs one part of salt (Kauffeld *et al*., 2005).

Layers of fish must be separated by layers of salt. It is a valuable method when one has no containers. This method is used to salt down flying fish in open fishing boats while at sea, and the fish in this case are kept whole. Some people like the salty taste of fish prepared in this way, but it is always possible to wash the salt away by soaking it in fresh water before use (FAO, 2005).

7.4 Drying

Very small and thin fish can be dried straight away in the sun if they are brought in early enough in the morning (and if, of course, the sun is shining). If these conditions are not fulfilled the fish must be put for one night in brine, or dry salted. They can then be dried the next morning (Deepchill, 2010). If it happens to be raining the next day, it is necessary to wait until the weather has cleared up, which could take from a few hours to a couple of days. In this latter case it will be necessary to wash the salt away from the fish by soaking it in fresh or sea water for a couple of hours before drying it; this depends again on the tastes of the consumers and on the purpose for which the fish is cured (Huss, 2009).

Small fish are mostly sun dried on mats, or suspended. When it rains the fish must be kept dry by covering or transferring them under shelter. If fish are laid on mats or other material to dry, it is best to turn them over every two hours so that they will dry quickly and not become maggotty. In the case of large fish, hanging is better if they are merely split (Ananou *et al*., 2007).

Dry salted fish can also be dried, but they should first be cleaned in water. Normally the fish will be dried after three days. If a great quantity of fish has been dried and is to be kept for some time, the best way is to pile it up in a dark place, off the ground and preferably on wooden boards. It should then be covered with a sack or mat.

After a fortnight the fish should again be laid in the sun for one or two hours and then put away as before. These are only indications of the main principles of fish drying; variations are possible (Leister and Gould, 2002).

7.5 Smoking

Any kind of fish can be smoked. There are three main methods of smoking:

(a) Smoking and roasting;
(b) hot smoking;
(c) long smoking.

•**Smoking and Roasting*:*** This is a simple method of preservation, for consumption either directly after curing or within twelve hours. Re-smoking and roasting can keep the product in good condition for a further twelve hours (Kauffeld *et al*., 2005). Fresh unsalted fish is put over a wood or coconut husk fire. This should be kept very small and the fish turned over every five minutes. In about half an hour the fish is ready for consumption or, if it is the intention to keep it for a while, it should be put in an aerated container (Tys and Pieters, 2009).

Fish can be preserved in this way even in open fishing boats, but the smoking has to be done in a tin or a half-drum. Salted fish can also be smoked by this method, but this is used mostly for immediate consumption or in order to bring the produce in smoked form to a nearby market.

•**Hot Smoking:** The hot smoking system can be used for immediate consumption or to keep the fish for a maximum of 48 hours. Small fish can be salted first for half an hour (see wet salting). After salting they are put on iron spits and dried in a windy place or in the sun for another half hour. It is necessary to have an oil drum to make the smoking stove. The top of the drum is cut out and holes are made 8 inches below the rim to place spits. Near the bottom a rectangular opening is made to control the fire. This opening should be closed with a small door or piece of steel plate. A fire of hardwood or coconut husks is made in the stove, and once it is well started it is regulated so as to give no flames (Tys and Pieters, 2009). The fish are then placed over the spits. During the smoking operations the top of the drum must be covered with a sack or with palm fronds laid as close together as possible; the fire control opening should also be closed. The fire must be watched from time to time. The fish will be ready in about one hour. An indication that they are done will be found in the golden yellow colour of the skin. For big fish, l i to 2 feet long, the best method is to split them in halves, to the right and left of the backbone. Each half fish is fixed between two flat bamboo slats or sticks. These halves are then rested head down on racks built four feet above ground. A number of split fish can be lined up next to each other.

A fire of hardwood or coconut husks, or several separate fires, are then lit under the rack. The number of fires depends on the quantity of fish one has to smoke. There should be a slow fire for about half an hour followed by a brisk one for one hour. A small fire is then kept going for six hours (just smoking) (Alasalvar *et al*., 2011).

After this treatment the fish is ready for transport and will keep in good condition for two to three days under tropical conditions. This method is used in particular in the Celebes for skipjack and other tunas (Ananou *et al*., 2007).

•**Long Smoking:** If fish must be kept in good condition for a long time, for instance, two or three months or even longer, it can be done by smoking, provided the fish is not oily.

For this purpose, a small closed shed made of palm leaves or other local material can be used. The dimensions of the shed depend, of course, on the quantities of fish to be smoked, but the height should in no case be less than six feet. In this shed, racks are built to hang the fish from or to lay them upon. Hanging the fish on spits is the best method, but they can also be laid on loosely-woven matting. One can start hanging fish three feet from the bottom up to the roof (Deepchill, 2010).

The preservation of fish is affected by smoke only in this method, and it is best to use coconut husks which should burn very slowly so that the fish is dry smoked after 48 hours. After such a treatment the flesh is dried throughout. If it is necessary to transport these fish to other islands, they should be packed in small packages wrapped in dry leaves and reinforced with bamboo or sticks. In Eastern Indonesia, packages of smoked fish are sent over great distances (Idachaba, 2001).

8. Fish canning

This is a process involving heat treatment of fish in sealed containers made of tin plates, aluminum cans or glass, until the product has been fully sterilized (Idachaba, 2001). During caning, heat treatment should be sufficient to destroy all heat sensitive bacterial and spores, in activate, the enzymes and cook the fish so that the product remains acceptable to the consumer after prolonged storage i.e (FAO, 2005) commercialized sterilization this is used in thermal processing to describe the heat treatment designed to kill substantially all microorganisms and spores which is present and cable of growing in the product (FAO, 2008). The canned food fish is also prevented from contamination by pathogenic organisms by storing them in a virtually airtight package. If heat treatment is properly carried out canned fish may remain in storage for several years without refrigeration (Leistner and Gould, 2002). Traditional canned fish are obtained from small pelagic fish species such as herrings (*Clupea spp*), Sardines (*Sardinella sp*), Mackerels (*Scomberomerus sp*), Anchovies (*Engraulis sp*), Tuna (*Thunnus sp*). Bonga (*Ethmalosa sp*) (Gopakumar, 2010). Fish intended for canning must be in first class condition and must be handle in hygienic manner to reduce microbial load on the fish. Poor quality fish will produce canned fish with offensive odour and flavour, poor texture (Burt, 2003).

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