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ARTIFICIAL BAIT ALTERNATIVES MAINLY BASED ON FISH WASTE

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Quality of Life and Management of Living Resources

ARTIFICIAL BAIT ALTERNATIVES MAINLY BASED ON FISH WASTE

ARTIBAIT

Individual progress report for the period
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Skýrsluágrip Rannsóknastofnunar fiskiðnaðarins
Icelandic Fisheries Laboratories Report Summary

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Ágrip á íslensku:	Þessi skýrsla er fyrsta framvinduskýrsla Rf í Evrópusambandsverkefninu: "Þróun beitu til línuveiða". Markmið verkefnisins er að þróa gervibeitu fyrir botnfiskveiðar með línu, aðallega á þorski, ýsu og lýsingi. Aðilar frá Spáni og Portúgal taka þátt í verkefninuog er takmark þeirra að þróa beitu fyrir lýsing. Stefnt er að því að beitan verði búin til úr afskurði frá fiskvinnslunni og/eða úr bræðslufiski og að í samanburði við náttúrulega beitu þá veiði hún álíka vel eða betur, verði á samkeppnishæfu verði og hæfi betur í vélbeitningu. Með vali á beituhráefni væri hugsanlega hægt að stýra hvaða tegund er veidd. Einnig er talið að stærð beitu geti að miklu leyti stjórnað stærð fiska á línunni. Í verkefninu hefur verið þróuð aðferð og smíðaður tækjabúnaður til að framleiða samsetta beitu úr frystu hráefni. Komið hefur í ljós að hökkun,hræring og tvífrysting virðist eyðileggja ferskleikaeinkenni beitunnar sem fiskurinn laðast að. Hins vegar hefur reynst vel að raspa frosnar fiskblokkir af beituhráefni, svo sem loðnu, sandsíli, smokkfiski, afskurði og setja fisksagið í litla trefjapoka. Skilyrðið er að hráefnið sé mjög ferskt þegar það er fryst. Beitunni er pakkað í trefjapoka sem henta vel til þess að stjórna útstreymi beituhráefnisins í nægilegu magni í æskilegan tíma. Efnið í trefjapokunum hefur einnig þá eiginleika að þegar það blotnar mýkist það og áferðin líkist þá helst roði á fiski. Pokabeitan hefur verið prófuð við raunverulegar aðstæður og lofar góðu.		
Lykilorð á íslensku:	*beita, gervibeita, línuveiðar, afskurður, bræðslufiskur*		

Skýrsluágrip Rannsóknastofnunar fiskiðnaðarins

Icelandic Fisheries Laboratories Report Summary

Summary in English:	This report is the first progress report from the IFL in the EU project "**Artificial bait alternatives**, **mainly based on fish waste** (Q5CR-2000-70427). The objective of the project is to develop an artificial bait, mainly for cod, haddock and hake, for the long-lining deep-sea fishing fleet. Spain and Portugal are participating in the project. The aim is that the bait will be made from fish waste from the fish processing plants and/or pelagic fish which is mainly utilized in fish meal production. In comparison with traditional bait, the developed bait will have a better or similar effectiveness, should be competitive in price and be more suitable for mechanised longlining. By choosing the bait material it will be possible to improve selectivity for fishing. In the project a method has been developed and equipment built to produce artificial bait from frozen material. It has been noticed that mincing, stirring and double freezing spoils the freshness characteristics of the bait which the fish is attracted to. On the other hand it has worked well to grate frozen fish-blocks of raw material like capelin, sand eel, squid and fish waste and put the grated fish material into small bags. It is essential that the raw material is very fresh when it is frozen. The bags are made from fibre that seem to be able to control the outflow and timing of the effluent from the bags. When the fibre material in the bags gets wet it softens and the texture feels like fish skin, which is a very positive property for a bait. The bait bags have been tried in real situations and are promising.
English keywords:	*bait, artificial bait, long-line, fish waste, pelagic fish*

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1 Objectives And Expected Achievements

1) To develop an artificial bait for commercial deepwater species, mainly based on waste from fish processing plants which
   - will have a better or similar effectiveness than natural bait
   - will have a competitive price compared with the natural bait
   - will be more adequate than natural bait for mechanised longlining
   - will be easily available
   - will improve selectivity for fishing
   - will preferably offer the possibility of storage without freezing

2) To define technically a factory dedicated to the production of the new bait and to study the feasibility of this new activity.

3) To study the repercussions of the new bait in the fields involved (fleet, processing plants and end-users).

The whole work will be carried out with an economic outlook, in a way to avoid solutions scientifically efficient, but economically unfeasible.

2 Project Workplan

2.1 Introduction

The primary goal of this project deals with the survival of the longlining deep-sea fishing fleet, at present in crisis because of the increasing costs, one of them baits. Because of the high running costs, some vessels have been withdrawn from fishing, and others are changing their fishing method towards trawling or gillnetting with the result of less selectivity and less environmental considerations.

In the second place, this project deals with the exploitation of by-products from the fishing industry, particularly from fish processing plants to elaborate artificial baits for the target fisheries. Some aspects from these fishing industries must be considered:

- Fish processing plants are usually situated in coastal areas and are directly connected with operating fisheries.
- Waste products from these industries give rise to environmental problems.
- The sector is going through a difficult time.

In the third place, the project will fit with the European fishing policy at present limiting the catches of sardines (*Sardina pilchardus)* and mackerel (*Scomber scombrus*) because of the overexploitation of the stocks (see EC Regl. Nº 2574/97 Com. of 15/12/1997, EC Regl. Nº 2429/96 Com. of 17/12/1999 and EC Regl. Nº 2968/95 Com. of 19.12.1995, funding the reduction of fishing quotas).

The decision leading to the development of artificial baits in general using by-products of fish processing plants contribute to fulfil the objectives of the Quality of Life and Management of Living Resources. The key action involved would be the 5^{th}: Sustainable Agriculture, Fisheries and Forestry, and Integrated Development of Rural Areas including Mountain Areas. Development of artificial baits using by-products will reduce the vulnerability of the fishing industry sector , will have implications for employment in coastal areas and finally, is a way of sustainable use of wastes, considered as a renewable resource, from those fish processing plants.

ICELANDIC FISHERIES LABORATORIES (**B3**) is an RTD performer which has obtained important experience in developing food for aquaculture and artificial bait and also have a relevant background in fishing products.

It is estimated that after finishing this project successfully, in the term of one to two years the results can be marketable. During this period, a full-scale test should be carried out and the technical design of the new activity will be completed.

The development of successful artificial bait for marine commercial species is an aim for many researchers dealing with chemoreception and will be welcome by fishermen organisations. Regarding hake, the knowledge about this species in this topic is very poor (Franco et al., 1987; Puente, 1995), as a gadoidiform fish is believed that its feeding behaviour is primarily mediated by chemosensory mechanisms. Although different types of substances are known to provoke responses in fish, chemosensory research involving behavioural and electro-physiological work have given strong support that free amino acids (specially L-stereoisomers) and other low molecular weight components of tissues are dominant in the aquatic environment in this respect. The stimulation of feeding behaviour is due in many cases to a mixture of chemical substances rather than a single one. The reason is because the appearance of synergistic effects between different substances. Taking into account this effect sometimes is not worthwhile to carry out long-term research to try to unmask a single attractant and only an effective fraction is explored. Visual stimuli (i.e. bait appearance, shape, brightness and size) in the longline catching process of hake are also an important factor that should be taken into consideration in the development of artificial bait for hake (Estepan). The feeding behaviour of the other target species has been studied more in detail, but so far artificial baiting has not been implemented.

The finding of substances that could act as attractant (a stimulus to which an animal responds by orienting toward the apparent source, possibly over long distances) and as stimulant (a stimulus that promotes ingestion and continuation of feeding) is the key factor in the development of an artificial bait. Other components in the artificial bait are a reinforcement (that gives physical strength to prevent bait loss) and a binder, which regulates the release of the attractant.

Preliminary research work (pers. com.) has given evidence that the best natural bait for hake is sardine (*Sardina pilchardus*) and this should be the starting point in the development of artificial bait for hake. Squid, sand eel, herring and mackerel are also important baits for other commercial species. These species are mainly caught for human consumption, and their price has reached unaffordable levels for the fishermen. Supply can also be a major problem for fishermen.

The aim of the present project will be the development of an artificial bait for hake, cod, and haddock, and other species such as ling, sea perch and pomfret, mainly based on wastes from fish processing plants and by catches (cepelin etc.) with similar free amino acids profile than the natural baits. Biochemical work will be followed by sea trials comparing the effectiveness of both artificial and natural baits.

In spite of the present high prices of the natural baits from coastal fisheries, importation of these species for bait has not solved the problem, due to the lower quality and mainly, in the case of sardines, because of the freezing method required, based on brine, at present used for the production of bait. Both the freezing and the distribution of this bait implies significant costs which could be avoided if the new bait could be stored and distributed within a refrigeration chain, as most of the vessels involved in these fisheries are dedicated to fresh fish, and therefore they do not deal with frozen products. The holds of the new vessels could also be simplified if there was no need to store a frozen bait. Because of these considerations, the possibility of an artificial bait to be stored at room temperature or chilled would be highly interesting, and will be an objective of this study.

The feasibility of the ideas explained in this project is at present high, as the aim is to find alternatives to a product, natural bait, whose price is increasing considerably. Besides, the development of the equipment for the food industry is given more chances to be considered under this project.

There are two approaches to develop artificial baits (Bjordal & Lökkeborg, 1996): using synthetic baits based on extracts or synthetic chemicals or using surplus or by-products from the fishing industry as inexpensive baits. This project will focus on the second one as the main raw material, although other refined substances like fish oil will also be considered.

Comparative fishing trials with the experimental longline (with natural and alternative baits) will be carried out during the different seasons. Due to the low catch rate for most of the target species it is expected that about 50 sets of about 2000 hooks will be required. Scientists will follow up on the trials on board of professional vessels, and also on a school vessel.

The trials will be carried out focusing on fisheries of special interest to the fleets of three countries: Spain (hake, pomfret and ling), Portugal (hake and sea perch), and Iceland (cod and haddock).

The evaluation of the catching efficiency of each type of bait will be carried out following the classical method of “alternating blocks” used in longline efficiency studies (Skeide et al., 1986; Huse and Ferno, 1990; Lokkeborg, 1990; Lokkeborg, 1991)

By experience, for the species and fishing grounds that we are dealing with, and the catch rates expected, usually about 2.000 hooks are set to detect significant differences.

Usually, catch data is not normally distributed and the variances are not homogenous, so non-parametric tests must be performed. The Kruskal-Wallis test is used to compare median catch rates per tub between fishing trials and different baits. The X^2 test is used to test differences in hooking position and a pair-wise test for comparison of proportion is used in the case where the X^2 is not possible due to frequencies being less than 5.

The two-sample T-test is used to compare mean sizes.

The number of hooks with the same bait per section of longline is also based on practical aspects related to working on board and recording of data. Usually, in order to minimise the interference with the normal fishing activities of the fishermen, alternate tubs (a longline unit of 150 hooks) with the same kind of bait are used.

3 Materials and methods for the chemical and biochemical analysis

3.1 Proximate analysis

The Biochemical and proximate analysis were all performed at the Icelandic Fisheries Laboratories. Following is a description of the analytical methods used at the Icelandic Fisheries Laboratories.

Protein. Ghb-e-AM-903. The sample is digested in sulphuric acid in presence of Copper as catalyst. There after the sample is placed in distillation unit, 2400 Kjeltec Auto Sampler System. The acid solution is made alkaline by a sodium hydroxide solution. The ammonia is distilled into boric acid and the acid is simultaneously titrated with diluted H_2SO_4. The nitrogen content is multiplied by the factor 6,25 to get %crude protein. *Ref. ISO 5983-1979.*

Water. Ghb-e-AM-904. The sample is heated in a heating oven at 103°C +/-2°C for four hours. Water corresponds to the weight loss. *Ref. ISO 6496 (1983).*

Ash. Ghb-e-AM-905. The sample is ashed at 550°C, and the residue is weighed.
Ref. ISO 5984-1978 (E).

Fat. **Ghb-e-AM-901a.** The sample is extracted with petroleum ether, boiling range 40-60°C. The extraction apparatus is 2050 Soxtec Avanti Automatic System.
Ref. AOCS Official Method Ba-3-38 with modifications according to Application note Tecator no AN 301.

Salt (NaCl). Ghb-e-AM-902c. Soluble chloride is extracted from the sample with water containing nitric acid. The chloride content of the solution is titrated with silver nitrate and the end point is determined potentiometrically. *Ref. AOAC 16th ed. 1995 no 976.18.*

3.2 Amino acids analysis

Apparatus

The chromatography system consists of Hewlett Packard (HP) 1050 series (gradient) pumping system, HP autosampler, Varian 9070 fluorescence detector, Croco-cil columnheater and HP Chemstation datahandling system. The column (150x4.6 mm) from

Hichrom, Reading UK, is packed with 3µm Spherisorb ODS-2 material and suitable guard columns of same packing material used.

Sample preparation

The samples (2-5g) are accurately weighed into centrifuge tubes and 20 ml of hydrochloric acid (0.1 N) and 1 ml of an internal standard (norvaline) added. After homogenization for 1 minute, using an Ultra-Turrax homogenizer, the samples are centrifuged at 15000 rev/min for 20 minutes. An aliquot (1 ml) of the supernatant is then diluted to 25 ml with distilled water before analysis.

Derivatization and separation

The derivative, with OPA (o-phthalaldehyde) as derivatization reagent, is made in the autosampler. Each sample is run 3 times and standards before a sample triplica. At least one blank is run.

The amino acid derivatives were separated by reversed phase chromatography using binary gradient elution at 25°C.

Solvent A: acetate/methanol/THF(tetrahydrofuran) pH 7,0 and B: methanol.

Flow rate 1ml/min. Excitation wavelength 338nm and emission 456nm.

For more detail on amino acids analysis (Bragadóttir, M. 2001)

4 Literature Research on Artificial bait

4.1 Introduction

By reading the literature it can be seen that preparing an effective bait for longline fishing (cod, haddock) is a complicated task. For the fish to swallow the bait and thus get hooked is preceded by a major investigation by the fish. Everything has to coincide; smell, texture and shape. The chemical stimuli attracts the fish to the area, then the bait is investigated for texture and shape. Sometimes the fish can take the whole thing in its mouth i.e. bait and hook and then spit it all out without getting hooked.

Our literature survey is concentrated on longline fishing for cod in Icelandic waters. The major feed for cod in Icelandic waters is capelin (*Mallotus villosus*) and shrimp (*P. borealis*). In attempt to formulate an artificial bait for this species the chemical stimulant that resembles cod's natural diet has to be take into account. Visual stimulant is very likely unimportant as the capelin does not give any phosphoric impact.

By going through the literature it can not be seen that an effective artificial bait is on the market. All kinds of projects have been tried in the trust of preparing an effective artificial bait, especially the ones that survive soaking for many hours without loosing all their feeding stimulants. In 1970 a literature study was made at the Icelandic Fisheries Laboratories on the possibility of using artificial baits. The literature research covered the years 1960- 1970 (Salómonsdóttir 1970). It was found out that a lot of work has been done, especially in developing phosphorescent plastic bait, but there were no positive results reported.

The impracticality thing with ordinary bait from cuts of herring for example, is that it only tolerates soaking for few hours before it becomes soggy and looses the smell stimuli to attract the fish to the line.

4.2 Literature

4.2.1 Acceptance of bait and feeding stimulants

Johnstone and Mackie 1986, discussed research that was done at the Marine Laboratory in Aberdeen: Initially research was designed to make observations employing underwater television of the behaviour of fish towards several conventional baits such as mussels and squid fished on a short section of small line. From these observations the process of fish capture during baited line fishing can be broadly divided into four stages. The first stage involves the orientation of fish from a distance and is termed "attraction". The second stage, termed "attack" or "bite", is the initiation of feeding involving taste and touch. Thirdly, "hooking" occurs when the hook engages in the fish during bait sampling or after acceptance and finally, "capture", when the hooked fish is landed on deck. In general the average catch rate of 15 cod per 100 baited hooks is a reasonable return from commercially baited small lines. The low rate of successful capture was particularly surprising in view of the activity observed around the baited hooks. Television observations of the responses of fish to artificial baits in the sea demonstrated some of the shortcomings. Baits which attracted fish, often did not stay on the hook sufficiently firmly to withstand repeated sampling or attack. The bait was usually taken and devoured but was often dislodged from the hook before being swallowed. It appears that, even if a bait attracts fish and the bait is taken, examination continues within the mouth. If the bait is not acceptable it is often rejected at this stage, complete with hook. The problem thus seems to be to create the correct texture and taste of bait which, once taken, will stimulate a swallowing response and thereby prevent rejection of the ingested hook. In fishing trial (Løkkeborg 1991) on torsk (*Brosme brosme*), ling *(Molva molva)*, cod *(Gadus morhua)*, and haddock *(Melanogrammus aeglaefinus)* were minced raw material were used as feeding stimulants and nylon bag was used as reinforcement. The texture of the nylon bag had a negative effect on the catching rate, most pronounced for cod and haddock. Since most fish eat a variety of natural food materials with a wide range of textures. It appears that the crucial thing in development of artificial bait is to find the right taste.

4.2.2 Bait lost and release rate of attractants

Immediately after the bait is dropped into the sea it starts to loose its catching ability. There are three major reasons involved: Bait is lost in handling, that is how the bait is hooked on and how the longline is put into the sea, pillage such as starfish, crabs and other creatures and at last decreased flow of feeding stimulants from the bait.

Mackie and Shelton 1972, Robert E. Muller and W.F. Van Heukelem 1986, and Løkkeborg 1989 are all unanimous about the importance of amino acids, not as a single substance but in combination with other organic compounds such as quaternary ammonium compounds, nucleosides and nucleotides, and organic acids. Løkkeborg 1990 (1) used a fluorometric detection to determine chemical flow from mackerel and an artificial bait based on shrimp extract and carrageenan, by estimating the release rate of amino acids. The results showed that the release rate of amino acids from both baits is at highest level in the first 1,5 hours and is followed by a much slower release rate the next 2 -24 hours. The release rate for this particular artificial bait is similar to the release rate of mackerel but the artificial bait declined slightly more rapidly. In artificial bait one should be able to control the rate of realise by using a different type of binding agent and supporting material. In above trial were Løkkeborg used foam rubber plus 2 % carregenan, similar release rate were experienced as for a natural bait. Løkkeborg and Johannessen (1992) studied the importance of chemical stimuli in bait fishing. In fishing trials for torsk (*Brosme brosme*) they compared the catching power of baits soaked in sea water prior to baiting with a fresh bait. Mackerel baits pre-soaked for 2, 4 and 24 hours gave 87%, 84% and 50% of the catch rate of fresh bait, respectively. Compared with fresh bait, all pre-soaked baits gave a significantly lower catch of torsk and the difference in catch rate increased with increasing time of pre-soaking. The bait loss was significantly lower for the pre-soaked bait than for fresh bait in all comparisons. To trigger bait intake, taste stimuli are probably of great importance. Fish responding to the bait without being hooked indicates that pre-soaking may have made the bait less attractive to predators. Fish caught on pre-soaked baits may have been attracted to the vicinity of the longline by the odour plume from fresh baits, thereby causing an underestimation of the difference in catching power. Bait soaked for 24 hours prior to baiting gave a much lower catch rate than bait pre-soaked for 2 and 4 hours. This indicates that the release of attractants after 24 hours of soaking has decreased to a level that influences the catching efficiency considerably.

4.2.3 Size of bait and reduced catch of under sized cod

Løkkeborg (1990) (2) did an experiment where he found out that artificial bait in longlining can be size selective for the catch i.e it reduces catch of under-sized cod (*Gadus morhua*). The bait he used was provided by Whitney Marine Laboratory, University of Florida. The baits were composed of a reinforced polyurethane foam impregnated with feeding attractants that occur in natural shrimp bait. No further details of the feed attractant was given. The foam was cut into three different sizes; small (35x20x2 mm), medium (35x20x5 mm) and large (50x20x8 mm). The concentration of attractants in each bait was normalised so that each bait released the same amount of attractants irrespective of size. The bait swell when soaked in water and the real sizes when fishing were, (35x21x4 mm), (39x24x9 mm) and (56x22x15 mm). The natural control bait was shrimp (*Pandalus borealis* Krøyer) of one size (mean 79x33x14 mm). The natural bait was therefore larger than the artificial bait and differed also in shape. The total catch rate ranged from 20 to 43 fish per 100 shrimp-baited hooks. The small artificial bait gave nearly the same catch rate as the natural shrimp bait, whereas artificial bait of larger sizes compared inferiority with the shrimp bait. In all comparison artificial bait caught a lower number of small cod, whereas there was no difference in the number of large cod. The proportion of under-sized cod (< 42 cm) caught on artificial bait was significantly lower than for natural shrimp bait. This difference cannot be explained by the effect of absolute bait size. Larger baits have been shown to catch less longer cod than smaller baits (McCracken 1963;Johannessen 1983). Also larger cod have a greater diet breadth (Mehl et al 1985) and therefore have greater experience of different prey types.

4.2.4 Material for making artificial bait

Løkkeborg (1991) did a fishing experiments with an alternative longline bait using surplus fish products. Bag-enclosed bait of minced herring gave higher catch rate for torsk, ling and haddock than natural bait.

Johnstone and Macke (1990) did a comparison of feeding stimulant activity of squid (*Loligo forbesi* Steenstup) a mantle ethanol extract, synthetic squid mixture and component of that mixture for juvenile cod. That ethanol extract of squid and synthetic squid mixture were eaten in equal amounts shows that all the chemicals

acting as feeding stimulants are present in the synthetic squid mixture and that lipids are not involved. The results demonstrate that L-amino acids are the major feeding stimulants for cod. The aromas, basic- and acidic amino acids were feeding deterrents. This study demonstrates also absolute stereospecificity at the receptors surface. L- neutral amino acids were less effective than the L- amino acid mixtures but D- neutral amino acids were inactive. In paper published Mackie (1973) were he is looking at feed stimulants for lobster (*Homarus gammarus*), his results indicate that a high degree of stereospecificity is involved. Mackie and Shelton (1972) showed that a number of single amino acids attract lobster (*Homarus americanus*) but that certain mixtures were more attractive than any of the amino acids individually. They also tried the attractiveness of the bait when lipids were removed from the filtered extract and found out that attractiveness was unimpaired, which indicates that oils and fats are not involved. Therefore the main concern in preparing an attractant should be on the amino acids and amines.

4.3 Discussion

For many years people have tried hard to make an artificial bait for long line fishing that fishes well and is easier to handle than the ordinary bait from cuts of herring or cuts of other raw fish. In Iceland people started to look seriously into making artificial bait around 1970 (Salómonsdóttir, 1970) and now, 27 years

later no prosperity has become of it neither in Iceland nor other countries as far as can be seen. The research that have been done on artificial bait and how fish respond to a baited hook have shown that the factors involved in the act of a fish getting hooked on a longline are many. The bait undergoes a major investigation before the fish accepts it and bites on the hook. The smell has to be tempting, strong and long lasting enough to attract the fish to the bait, the texture has to be the right one and the bait has to have an acceptable taste. If the fish does not accept the taste of the bait when nibbling into it, the bait is not swallowed and the fish of course does not get hooked.

It is certain that a lot of investigations and testing are not reputed in the literature. Even though a lot of brainstorming, mixing and testing have been done on artificial bait, there is still no known artificial bait on the market today. Yet, each research project brings us nearer to the end point and that is making artificial bait that fish will bite on.

5 Research activities during the first reporting period carried out by partner B3

5.1 State of the art of artificial bait for target species - WP1

By reading the literature it was discovered that a lot of speculation and trials had been done on how to make a successful bait. As can be read in the literature report (Appendix 2) on the state of art of artificial bait experiments it is noticed that a lot of work has been done through the years in order to produce a successful bait for both long line fisheries and trap fishing. Some scientists aimed on the chemical stimuli of the fish by spraying for example a piece of foam with a certain mixture of amino acids. Other tried to mimic traditional bait by mincing waste from fish processing plants and underutilized fish species and use some kind of binders to get a specific texture of the bait. Different reinforcement were also tried for the fish mince for example, gauze, cheese cloth, nylon socks and all kinds of sausage skins. After the literature survey a good idea was obtained on how the artificial bait production was standing.

5.2 Market and biochemical analysis of by-products -WP2

5.2.1 Raw material available

A survey was done in Iceland to identify the sources and availability of fish waste from processing plants, underutilized fish species, waste from shellfish factories etc. which can be used as raw material for bait production. Table 1 demonstrates the results. The waste raw material is free of charge and the only cost is the transport.

Table 1. Results from the market survey on the availability and prices of fish and shellfish waste and species that go into fish meal production and can be used in artificial bait production.

Fish waste and under utilized species	Metric tons/year	Availability	Cost (Transport Km in averages)
Capelin	50.000 (ca 5% of annual catch)	July to March	€ 12 / Metric ton
Blue whiting	10.000 " " " "	Most time of the year	€ 20 / Metric ton
Cod waste	30.000	All year	just transport (200 Km)
Scallop waste	3.200	Sept to Feb	just transport (100 Km)
Iceland Cybrine waste	80.000	All year	just transport (500 Km)
Herring waste	21.000	July to Feb	just transport (300 Km)

There is a tradition in Iceland, for large scale fish meal production from capelin, blue whiting and herring. Most of the capelin catch goes into fish meal production except for about 25.000 tons which are frozen into blocks and exported. Last year the capelin catch was 1.000.000 metric tons and blue whiting 200.000 metric tons. If these species could be used in bait production it would make tremendous capital goods for the fishery sector. As it is today it is very difficult to use whole capelin as bait because the skin is very thin and weak so the fish falls easily of the hooks. It would be very important to be able to do some kind of preparation on the capelin, like mincing it or grating so it could be used as bait. On of the main prey for cod in the North is capelin so it should be a successful bait. The most popular bait for long-liners in Iceland are squid and sand eel but they are both imported to Iceland.

5.2.2 Chemical analysis

Proximate analysis of protein, water, fat, salt and ash was performed at IFL (see Appendix 1 for materials and methods) on all possible raw material available for alternative bait production as well as all types of traditional bait used in Iceland.

Table 2. Results from proximate analysis of traditional fish baits and possible bait alternatives

Traditional baits and alternatives	Protein (%)	Water (%)	Fat (%)	Salt (%)	Ash (%)
Squid	18,0	76,1	2,4	1,1	1,9
Sandeel (caught in Sept)	17,5	70,1	9,0	0,4	2,3
Herring (caught in April)	15,6	59,6	21,9	0,2	2,2
Mackerel	15,4	57,0	24,1	0,4	2,4
Capelin (caught in February)	14,1	74,8	8,7	0,4	2,0
Iceland cybrine	16,2	75,6	1,4	1,5	2,5
Norbait	14,4	71,7	9,0	0,5	2,3
Common whelk	19,4	75,2	0,3	1,1	1,8
Cod waste (fish gutted)	16,4	82,8	0,1	0,3	1,2
Herring waste	14,4	66,6	16,0	0,7	3,2
Scallop waste	12,5	85,7	0,2	0,3	0,7
Iceland cybrine (waste)	12,9	84,2	0,7	0,3	0,9
Blue whiting	18,1	80,5	0,8	0,1	1,2
Carpmeal	65,2	5,7	7,2	2,0	20,2
Blue whiting meal	72,5	7,8	7,3	2,0	12,7

Table 1 shows the results from the proximate analysis of traditional bait like squid, sand eel, mackerel and herring**.** The analysis included also fish waste from cod and herring, shellfish waste from the scallop and Iceland cybrine processing plants, capelin and blue whiting from the fish meal industry and fish meal from carp (bone meal) and blue whiting meal. Some fishermen in Iceland believe in common welk as a good bait so it was decided to look at the shellfish too. It is very interesting to see the analysis on squid which is the most popular bait in the long line fisheries in Iceland. The squid is very low in fat and high in protein so it is hardly stimuli from the fat that is attracting the fish to the squid bait more likely attractants from the protein. The Norbaits proximate analysis is very close to the analyses on the sand eel, maybe the Norwegian were imitating sand eel when they formulated the Norbait.

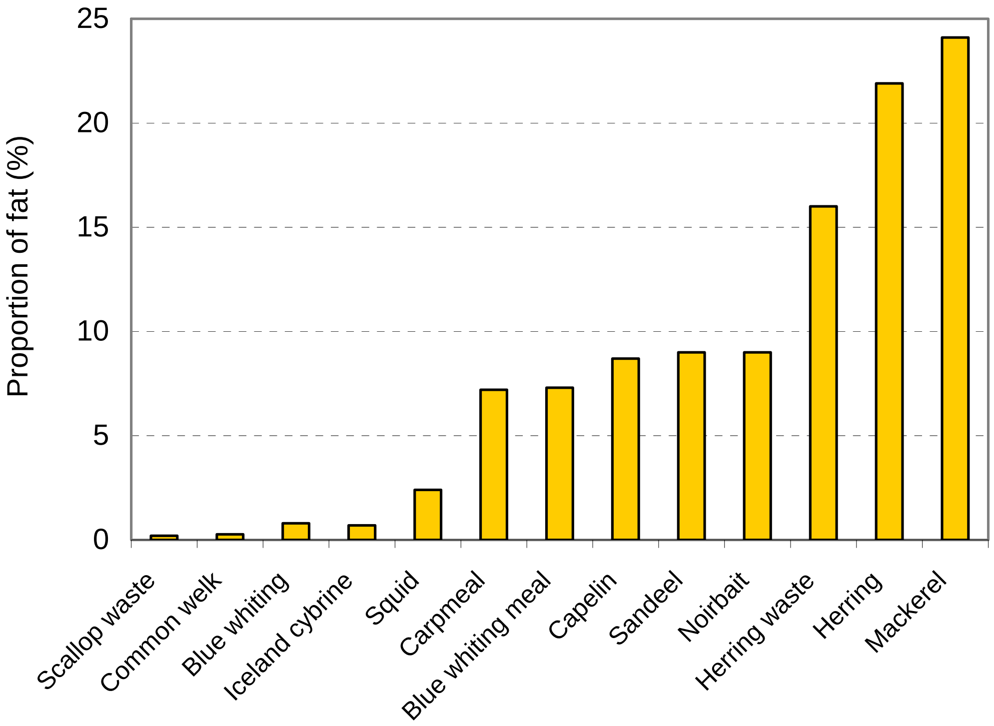

Figure 1. Fat (%) in various baits and bait alternatives from table 1

The different fat content in the different raw material is very evident in figure 1. Some of the raw material has hardly any fat content while other contain almost 25% fat like the mackerel. Mackerel is very important bait for haddock but not for cod. Haddock seems to take more bait with high fat content than cod. Haddock is also not as particular about the freshness of the bait as cod probably because haddock is a scavenger while cod only feeds on live prey.

5.2.3 Fatty acids analysis

Figure 2 shows the combination of saturated, monounsaturated an polyunsaturated fatty acids in traditional bait and the Norbait. Again the Norbait is very close to the sand eels combination in fatty acids as was seen for the proximate analysis. The polyunsaturated fatty acids give the most fishy smell of the fatty acids so it is expected that they would have the most attraction of the fatty acids. It can be seen that though the squid is very low in total fat content the proportion of polyunsaturates is quite high which very likely come from the cell walls as pospholipids. The proportion of polyunsaturates in the sand eel and capelin fat is also quite high.

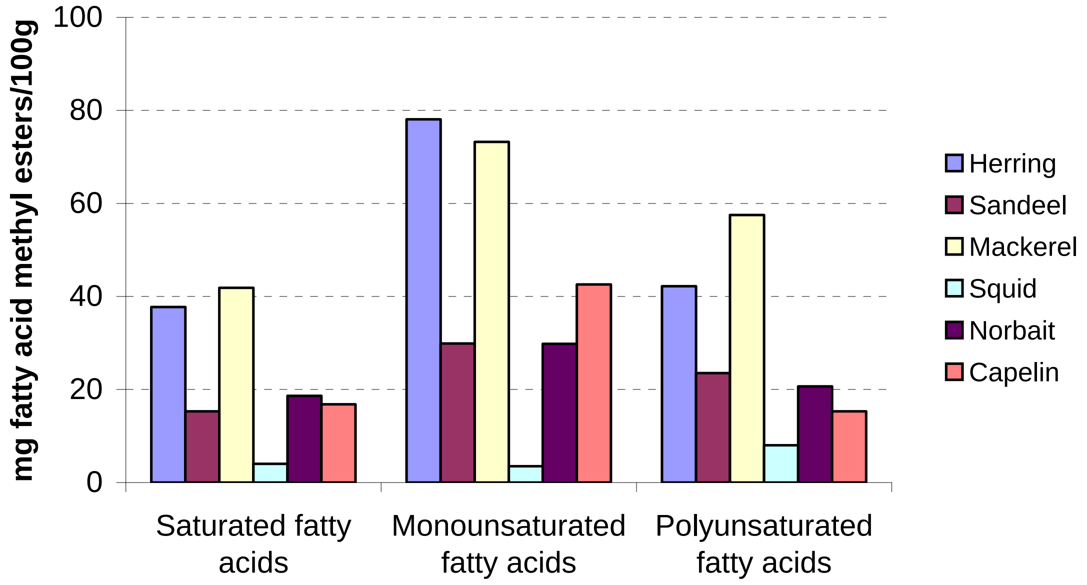

Figure 2. Combination of fatty acids (mg/100g)

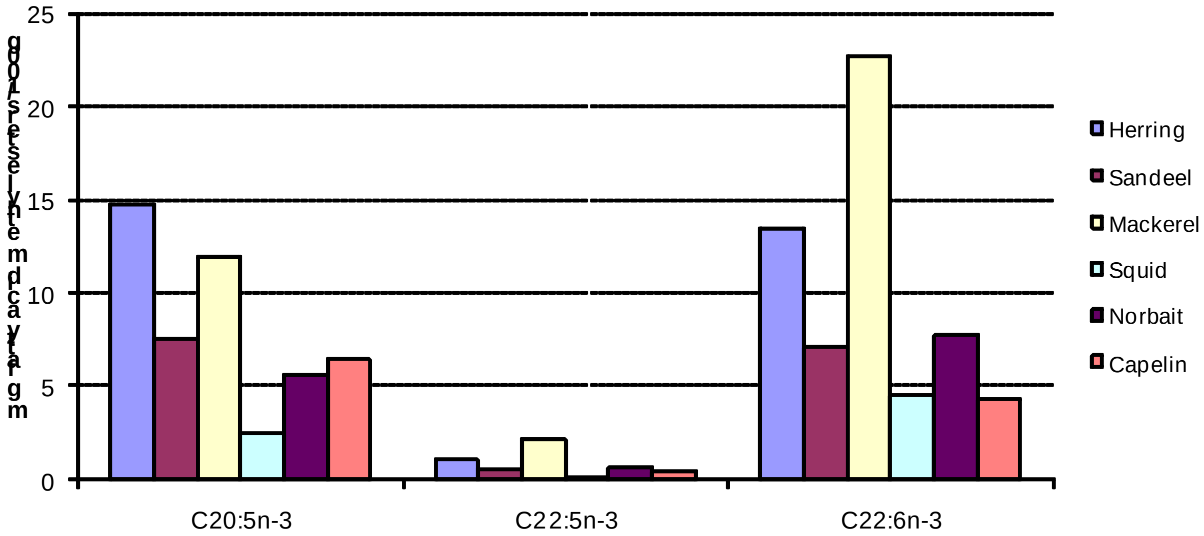

Figure 3. Polyunsaturated fatty acids

Figure 3 shows individual fatty acids of the polyunsaturated fatty acids group. The fatty acids from the raw material spreads quite evenly between the individual fatty acids. The proportion of the fatty acids in mackerel and squid is highest in the most unsaturated fatty acid C22:6n-3 or DHA.

5.2.4 Amino acids analysis

Following is a list of the free amino acids that were measured in the traditional bait and bait alternatives (the abbreviations for each amino acid that are used in text and graphs are also listed):

Aspartic acid	**ASP**	Arginine	**ARG**
Glutamic acid	**GLU**	Taurine	**TAU**
Asparagine	**ASN**	Tyrosine	**TYR**
Serine	**SER**	Methionine	**MET**
Glutamine	**GLN**	Valine	**VAL**
Histidine	**HIS**	Phenylalanin	**PHE**
Glycine	**GLY**	Isoleucin	**ILE**
Threonine	**THR**	Leucin	**LEU**
		Lysin	**LYS**

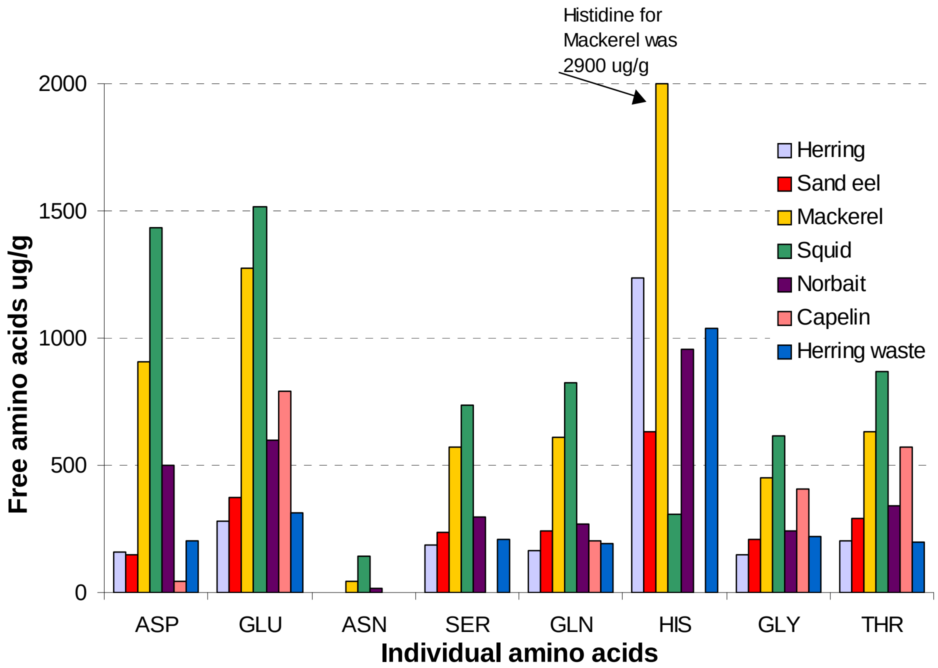

Figure 4. Free amino acids measurement of eight amino acids (Asp,Glu, Asn, Ser, Gln, His, Gly, Thr) measured in traditional bait.

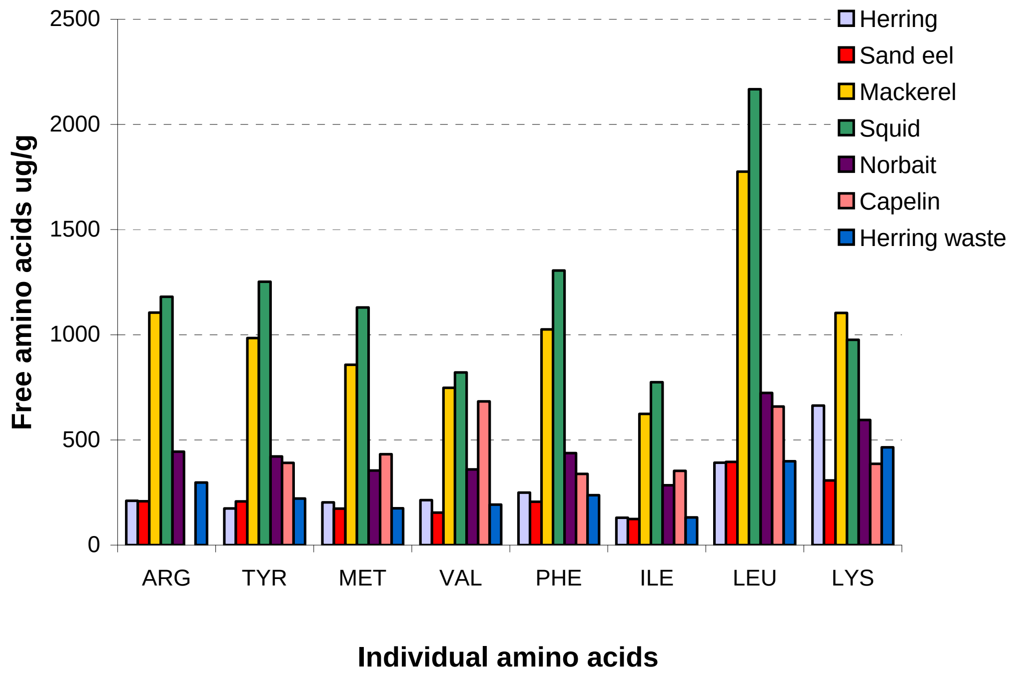

Figure 5. Results from free amino acids measurements of Arg, Tyr, Met, Val, Phe, Ile, Leu, Lys in traditional bait.

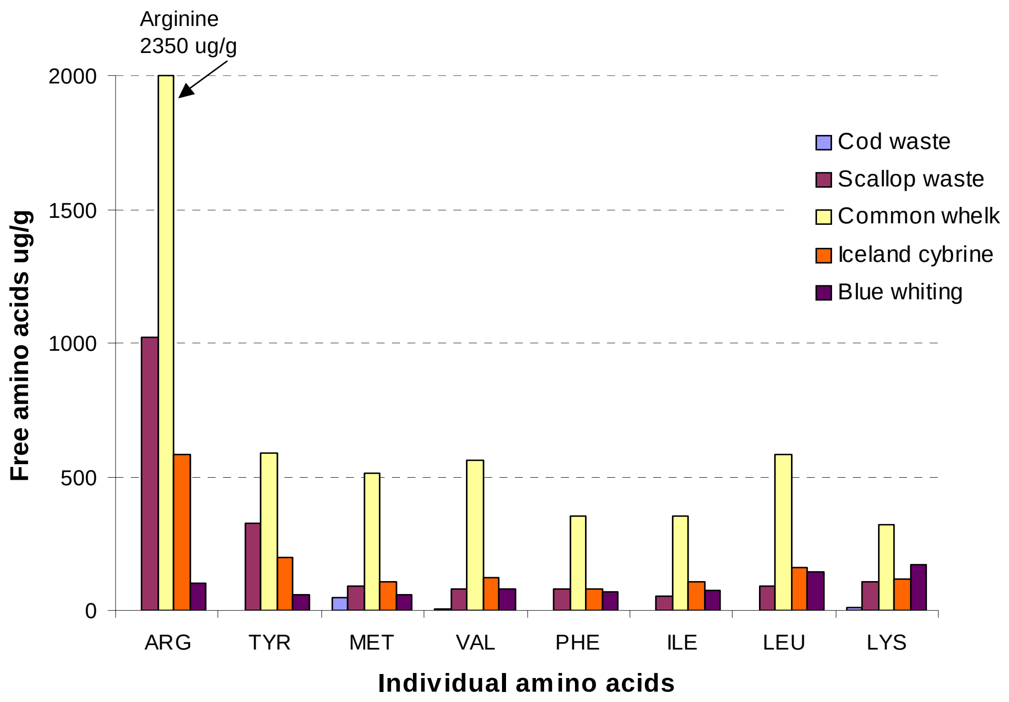

Figure 6. Results from free amino acids measurements of Arg, Tyr, Met, Val, Phe, Ile, Leu, Lys in possible bait alternatives

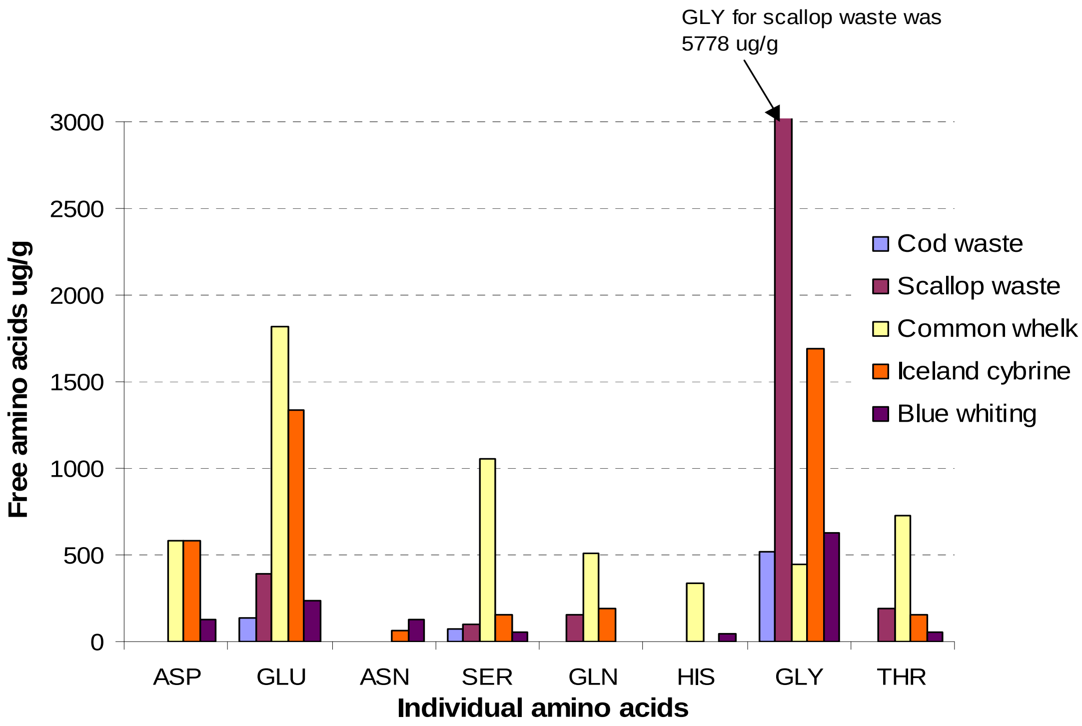

Figure 7. Results from free amino acids (Asp, Glu, Asn, Ser, Gln, His, Gly Thr) measurements in bait alternatives of fish waste and underutilized fish specie (blue whiting).

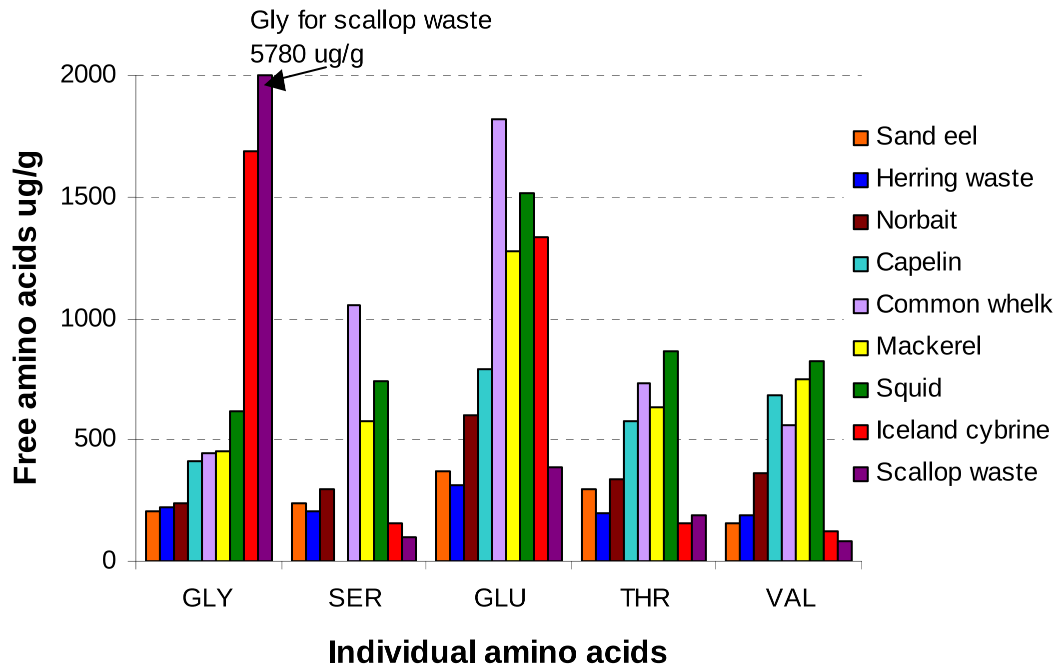

Figure 8. The five free amino acids (Gly, Ser, Glu, Thr, Val) that are believed to evoke food search behavior in cod (Hara 1992). Amount shown in different traditional baits and bait alternatives.

The amino acids analysis have been performed on traditional bait and bait alternatives and the values are shown in figure 4 to 7. A thorough study on the connections between individual amino acids and different bait materials has not been completed yet. In figure 8 the five amino acids which are thought to be the most effective amino acids as attractants for fish are shown. It can be seen that squid is very high in four out of five of the amino acids. The shellfish and shellfish waste have quite high values for these five particular amino acids. It is interesting that herring waste, sand eel and Norbait have the lowest values in all of the five amino acids. It is unfortunate that sandeel has this low values as it is a popular bait for cod. This weakens the free amino acids theory on potent attractants, but of course this remains to be studied further.

5.2.5 Data analysis

Principal component analysis (PCA) was done to study the main trend in the data. The PCA plot illustrates the similarities and dissimilarities in the composition of the raw material. Figure 9 shows a PCA biplot of proximate analysis data of various bait raw material. Mackerel, herring and herring waste were described by higher fat content and lies close to each other on the left side of the plot. Water content was negatively correlated with the fat content, i.e. with lower fat content the water content was higher as seen with e.g. cod waste. Samples lying on the upper part of the biplot, especially cybrine, whelk and squid, were described with higher content of salt, protein and ash.

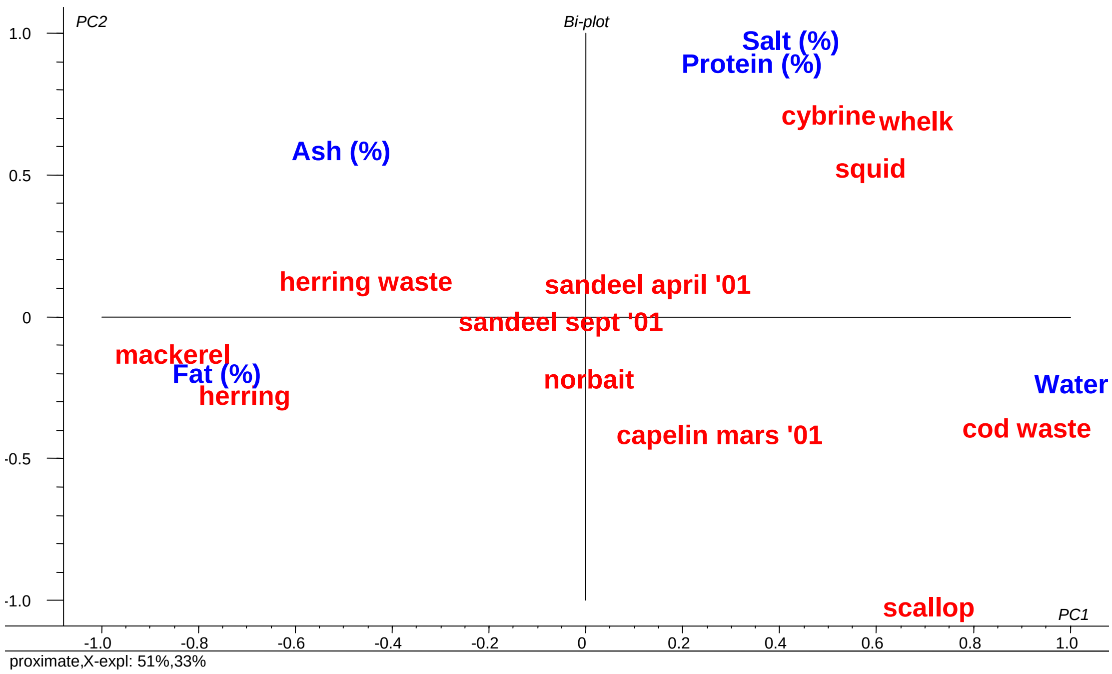

Figure 9. PCA biplot of proximate analysis data in various bait raw material

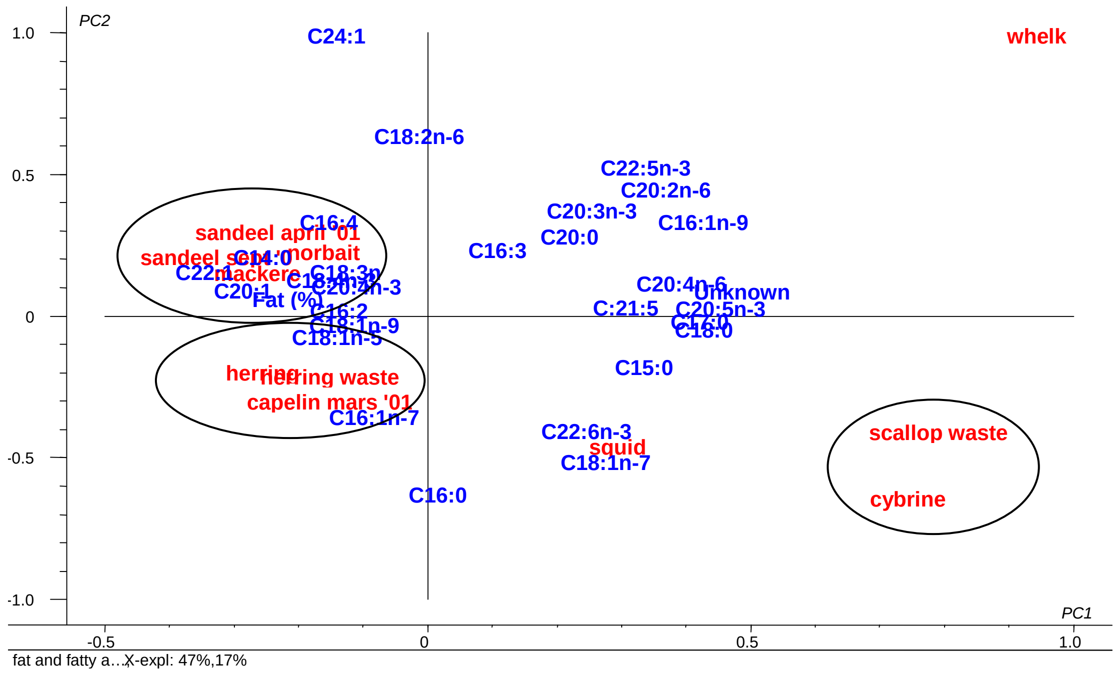

Figure 10. PCA biplot of fat and fatty acid composition in raw material

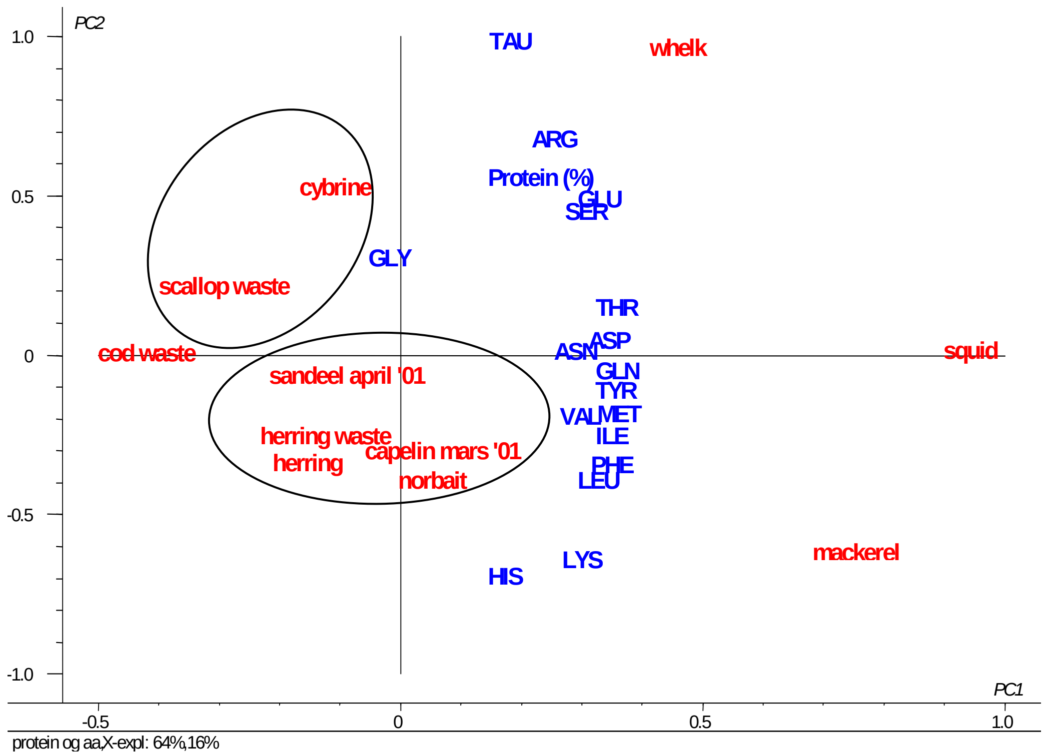

Figure 11. PCA biplot of protein and amino acids composition

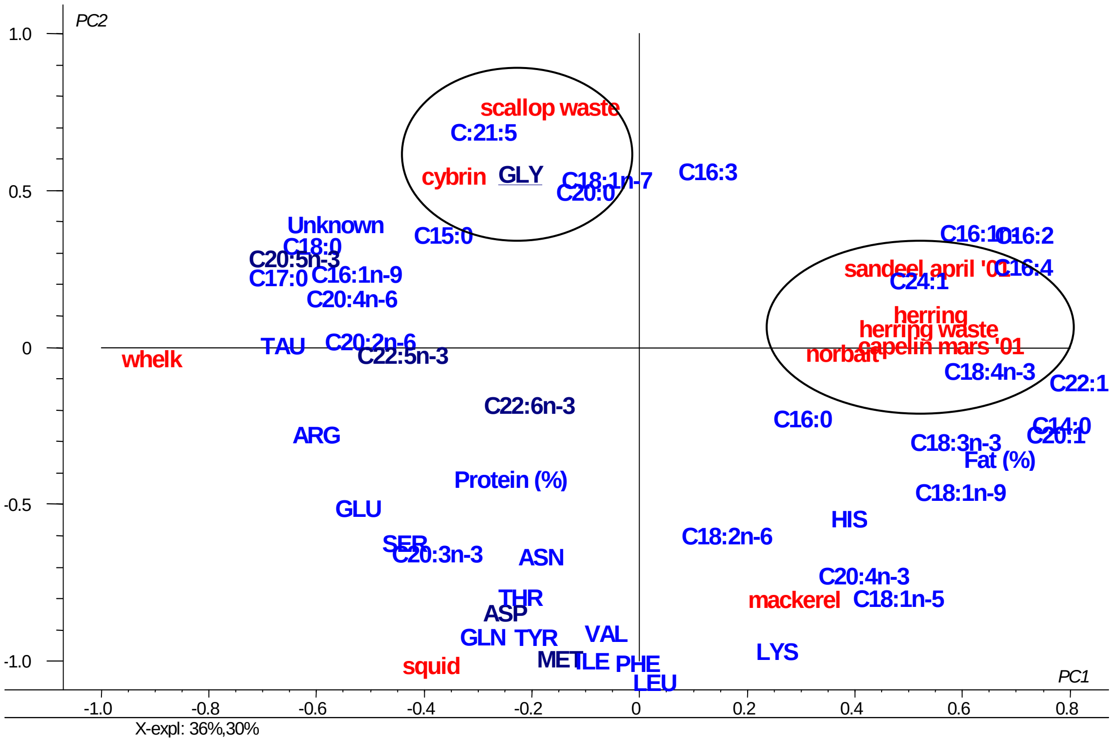

Figure 12. PCA biplot of protein , amino acids, fat and fatty acids

Figure 10 shows PCA biplot of fat and fatty acid composition in raw material. Samples with high fat content, like sandeel, mackerel and herring lie together on the left side of the plot and had similar fatty acid composition. They were described with relatively higher content of monounsaturated fatty acids. Some differences could be seen between these samples as emphasized with the two circles (Fig.7) and described by PC 2. The difference was mainly in the content of C24:1 vs. C16:0.

Samples with low fat lies on the right side of the plot and had relatively higher content of polyunsaturated fatty acids and unknown. These are samples like whelk, scallop waste and cybrine. Scallop waste and cybrine had similar fatty acid composition but whelk was unlike all the other samples in fatty acid composition.

Figure 11 shows PCA biplot of protein and amino acid composition of the raw material. Samples with higher content of amino acids, i.e. whelk, squid and mackerel, lie on the right side of the plot. As with the fatty acid composition whelk is unlike all the other samples with higher protein content and different amino acid composition. It is of interest to see that mackerel has totally different amino acid composition than sandeel and norbait but these samples had similar fatty acid composition. On the other hand the amino acid composition of scallop waste and cybrine is more similar to the samples with high fat content although they had different fatty acid composition.

In Figure 12 all the results are but together in a PCA biplot, i.e. fat and protein content, fatty acid and amino acid compositions. Some samples were left out because of lacking data. The first two principal components explained 66% of the variance in the data set, 36% by PC1 and 30% by PC2. As can be seen on the figure sandeel, herring, herring waste, capelin and norbait had a similar profile of fatty acids and amino acids. The fat content and the amount of monounsaturated fatty acid was high. The content of C18:4n-3, C18:3n-3 and C16 fatty acids were also high in these samples. On the opposite side of the plot are the samples with higher protein and amino acid content and low fat content. They were also described by higher content of long chain polyunsaturated fatty acids. Squid had also different profile compared to the all the other samples. Scallop waste and cybrine had a similar profile and lie close together on the upper part of the biplot. They had very high content of glycine, espesially the scallop waste as could be seen in Fig. 8. Glycine is one of the five amino acids that are believed to evoke food search behaviour in cod (Hara, 1992). Mackerel had similar fatty acid composition as the other fatty species (Fig. 2,3) but different amino acid composition (Fig.5,6). In Figure 5 it can be seen that the difference lies in higher content of lysin and especially in histidine that was very high in mackerel or 2900 µg/g (Fig. 3).

5.2.6 Preparation of the artificial bait-WP3

After an experimental period on formulating an artificial bait it was found out that thawing, mincing, mixing, stirring and re-freezing the bait material reduces the effect of the attractants that give the necessary freshness (chemical) stimuli that attracts the target fish to the bait. Cod is especially sensitive to freshness which is not surprising as it feeds only on life prey. It was very important in the beginning to thoroughly examine the paper reinforcement. To do that we had to use traditional bait and compare in sea trials with the same bait hooked traditionally and the same kind of bait minced and packed into the paper bags. We worked mostly with sand eel and squid. Many bait mixtures were made and they tried at sea with different results.

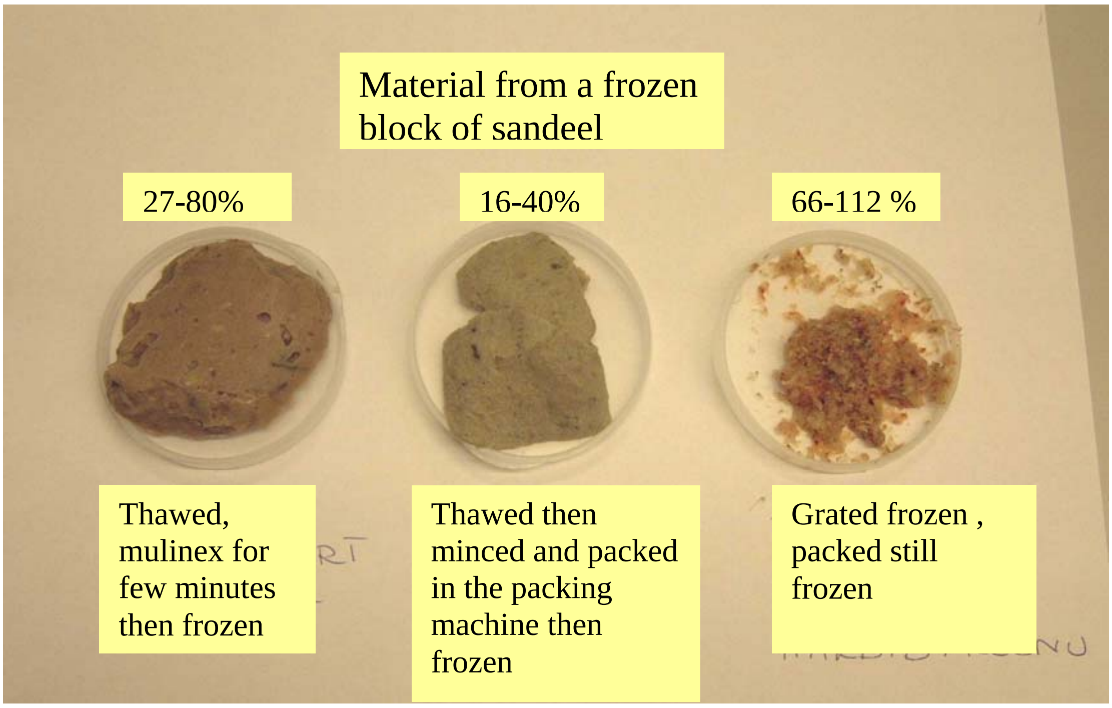

Figure 13. Different bait preparation of the same material

We noticed that if the prepared bait was taken to a sea trial right after it had been prepared it gave a reasonable catch but when it had to be kept in a freezer it was less and less effective the longer it was kept in the freezer. It should be noted that the bait was wrapped in plastic during the storage in the freezer. At this point it was noticed that the colour of the raw material changed a lot from transparent yellowish colour of fresh fish to greyish-brown when it came out of the grinder and packing machine and when it was kept in the freezer it always went more greyish and had less fishy smell to it the longer it stayed in the freezer.

Figure 13 demonstrates the effect a different handling has on the raw material.

Before the picture was taken the different baits had been standing in room temperature for 30 minutes. The raw material in the bait on the left was put in Mulinex for few minutes. It got this brown colour but it looks quite fresh. The catch was 27-80% compared to traditional bait. Most of the catch was haddock and catfish. The bait in the middle went through a the packing machine process i.e. thawing, mincing and packing. It is obvious that this handling changes the raw material. The bait has got a greyish mat appearance. In the sea trial this bait caught only 16-40% of what the traditional bait caught. All of the catch was haddock and catfish. The bait to the right is the bait that is grated frozen and kept frozen the whole process. It can be seen from the picture that the material has a transparent beige colour indicating freshness. The blood and material from the viscera can easily be detected.

This bait caught 66-112 % of what the traditional bait caught and moreover the catch was 60 % cod. This bait looks very promising. Now we are waiting for a grating machine that Dimon is developing and constructing. The machine will be ready in May 02. Dimon has also purchased a small packing machine that will be easy on the raw material. The machines have to be placed in a freezer.

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