#### Live Handling and Transport of Crustacean Shellfish: An Investigation of Mortalities

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SR280 MAFF R&D Commission 1985/86

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#### Summary:

This report describes the most recent in a series of investigations of mortality in crustacean shellfish during the export process. The main species involved is the brown crab *Cancer pagurus*. The complete sequence was examined from catching in the Spain and France. Data collected onwater quality and animal physiology were related and a number of factors were identified as contributing to the mortality problem. Thermal shock on reception abroad is seen as the most likely cause of death although more in terms of being the final straw than the main causative factor.

A number of recommendations are made to upgrade the export system at each stage: pre-sale holding, transport and final reception. Further recommendations are for a survey of the Western Isles stock of *C. pagurus* and for the development of a novel, integrated transport system.

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#### 1. Introduction and Background

The investigations described in this report are the most recent of a series of studies aimed at solving problems associated with the transport and marketing of live crustaceans such as crab.

The context within which these studies have been initiated by SFIA, is that of an expanding industry in a state of some uncertainty. The UK crab fishery is prosecuted in a number of regions and is based on several sub-stocks of the target species. The market opportunities for crab and for crab products are also distinct and the various regional fisheries have developed with traditional specialities in certain market sectors.

Now, however, there is a universal tendency for any producer to try to broaden the market appeal of his products. These moves have introduced a number of problems to the crab industry and it is to these that SFIA has addressed itself. The aim, overall, is to give each of the major regional fisheries the same chance of having a broad-based and demand-led stimulus to its development.

This particular series of studies started in 1983/84 with the development of a live trade in velvet crab (*Necora puber*) from Scotland to continental Europe which was plagued with heavy mortalities. SFIA commissioned a study by the University of Hull to find the underlying reasons and establish a methodology for such investigations.

That study started just as the Hebridean fishery for the brown crab (*Cancer pagurus*) was diversifying from pure processing into Live export. With that development came further, inexplicable mortalities which were all the more puzzling because they were occuring with companies which had a long and successful history of transporting English brown crab alive and in good condition to both home and continental markets.

The significance of excess mortality in this context is that it devalues the commodity in two ways. First there is a straightforward cash Loss equivalent to the percentage mortality; secondly the region of production starts to acquire a poor reputation and foreign buyers will start paying Less and Looking for alternative supplies. Already in 1985 there has been a significant Loss of markets by producers in the Hebrides as a result of a series of instances of mass mortality in their consignments to Europe. The countries involved include Spain, France and Holland.

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#### 2. Objectives

The following objectives were defined:

- i) to examine the phenomenon of excessive transport mortality of *C. pagurus* from the W. Isles and determine its cause(s).
- ii) to develop instrumentation which would enable water quality parameters to be monitored in transit.
- iii) to devise a development strategy for this sector of the crab industry.

#### 3. The Approach

As a result of the difficulties described in the previous section, SFIA once again contracted the University of Hull to carry out a biologically-based investigation of the phenomena. At the same time the electronics department of the SFIA's Industrial Development Unit devised a computer-based data Logging system which could record, then Later be used to control, the water quality in the crab holding tanks during transit. The parameters measured were dissolved oxygen, temperature, salinity and ammonia.

A third aspect of the SFIA approach was the employment of a consultant to make a thorough examination of first handling and storage practices by crab fishermen in the Western Isles. This was to check that there was no fundamental problem at this stage which was predisposing the crab to stress and subsequent mortality.

Broadly, the methodology of previous studies was used. This involved monitoring the handling sequence and recording the environmental conditions under which crab were held during each stage of their transport to Spain. These conditions were then simulated in the indicators of the crabs' physical condition ventilation rates, blood protein levels, Lactate and ammonia production. Crabs were also sacrificed at each stage of the transport process and post-mortems carried out. This enabled precise analyses to be made of their physical state at each point thus providing clues as to their reactions to these variations in their environment.

On the basis of work carried out previously it seemed likely that a combination of factors was effecting mortality none of which would necessarily have been lethal on its own. In order to locate the various sources of stress it was necessary, in the first instance, to characterise the events and influences within the catching, transporting and marketing system.

As far as Locations are concerned, there are four stages in the transport process:

- i) from time of capture until first sale to the exporter
- ii) whilst with the exporter until time of shipment
- iii) during transport
- iv) whilst with the importer

Of those factors which might have some influence the following can be identified:

- i) poor handling, selection and exposure at sea
- ii) prolonged storage without feeding
- iii) quality/condition of the crab
- iv) storage conditions ashore and during road transport
- v) length of journey
- vi) changes in water quality and lack of control over quality
- vii) bad handling practices on the Continent

With this rationale in mind these studies were designed with the intention of isolating as many of the above variables as possible.

A number of photographs illustrating the various holding facilities used and other factors are contained in Appendix II.

Reports from exporters suggest that the greatest losses occur during the summer months although, because the fishery is new, seasonality is not yet confirmed as a true part of the pattern. Also it seems that on the majority of occasions crabs arrive on the continent alive but large numbers then die at the importers' premises over the following 72 hours or so.

Exporters are also firmly of the opinion that Scottish crab are generally more susceptible to stress than English crab are. So it is apt to consider the possible differences that may exist between the exploited populations other than those imposed by the transport regime. These are the differences that would be loosely described by exporters as 'condition' though without any definition being applied to this term. It is considered likely that the English and Scottish crab populations may have condition differences as a result of environmental differences arising from their geographical location. Although these may not be particularly large they may be sufficient to act as crucial modifying influences on the respective tolerances of the crabs to the stresses of holding and transport. In the experiments attempts were made to equate 'condition' with the levels of various bodily functions of crab.

#### 4. The Examination of First Handling and Holding Practices

Dr E. Edwards, the consultant, spent several days in the Western Isles looking at shipboard handling and sorting techniques, the use of holding facilities like store pots and pounds and the process of grading and packing on to the final transport.

The visit was concentrated on the island of N. Uist with the help of the Barra Fishermen's Cooperative. This Coop has the advantage of having a very experienced English Channel crab-fisherman among their number. As a result they are aware of good handling practice and the benefits to be gained from it.

One consignment of brown crab was monitored from the point of catching through to despatch to France. Mortality levels were noted both on arrival and after a few days' storage at their destination.

The consignment, of one tonne, comprised 90% hen crabs and of these about 60% were at the point of spawning or immediately post-spawning. The crab was handled well at all stages and appeared lively and healthy on despatch. On arrival at their destination, however, mortality was about 50% and this figure increased to 70% after two days at the importer's premises according to information provided by the importers.

No measurements were taken of the water quality in the vivier tanks during the journey but the procedure followed was the same as on all other occasions. Under these circumstances the consultant deduced that the most likely cause of the high mortality rate was a 'condition' factor, probably related to the breeding cycle. It was noted that the proportion of hens at an almost identical stage of the spawning process was unusually high for brown crab and that this may be a characteristic of the stock around the W. Isles.

#### 5. The Biological Investigations

#### 5.1. Background

As noted earlier, these studies were framed around the need to monitor the crabs and their environmental conditions through each of the major stages of the chain of transport.

After they are caught, brown crabs' claws are usually 'nicked' - have their tendons cut - and kept in any of a number of storage pots or tanks for periods which may extend to 14 or more days before they are sold. Once nicked they can no longer use their claws to damage each other. There then follows a fairly short period when they are held by the exporter followed by the actual shipment to the continent - a journey time of about 2 days. During all this time the animals are not fed. This reduces costs and has the practical advantage of reducing the fouling of water during the risky periods of storage and shipment. However, a number of physiological changes may be expected to accompany extended periods of food deprivation and these will include uptake levels altering and blood protein levels dropping.

#### 5.2. The Experiments

#### 5.2.1. Oxygen uptake during periods of food deprivation

When a hen crab spawns her fertilised eggs she then glues the eggs to her abdominal limbs or apron, carrying them for weeks until they are ready to hatch. A hen crab carrying eggs is known as 'berried' and when in this stage she will stop eating and hide until her eggs hatch. This condition, however, is preceeded by an anticipatory build-up of food reserves which are utilised during the fast. At other times, if a brown crab is deprived of food it also shows a reduction of movements - presumably as an energyconservation strategy. At such times the animal's rate of use of oxygen declines rapidly during the first 7 days of deprivation and this is followed by a more gradual decline as starvation continues. Oxygen use in this context is an indicator of the amount of food an animal is using to maintain itself. In the absence of any external food source it must of course use its bodily reserves.

In laboratory experiments a general summer water temperature of 16°C was used at which to compare the oxygenuptake performances of 'settled' crabs deprived of food for 3 days and those after 10 days without food. The data are given in Table 1 and show that a drop of 18% in mean oxygen uptake took place between days 3 and 10. Normally, if allowed to do so, such animals spend Long periods immobile in the aquaria and remain spaced apart from each other. However, during storage and shipment animals are seldom, if ever, likely to become settled as they are subjected to such stresses as handling and packing, high density conditions and abnormally high levels of noise and vibration. Such events may be anticipated to promote elevated oxygen consumption rates just at a time when the animals should be conserving energy. Being realistic, it is difficult to envisage any suitable alternative handling procedures, apart from commonsense awareness of what is stressful and minimising this wherever possible. Clearly, the animals have to be kept and shipped at high densities for practical reasons and feeding, in addition to being an extra cost, does carry with it the risk of fouling occurring. Nor is it suggested that food deprivation is the principal cause of mortalities. It may be a contributory factor in this fishery but such practices are commonly employed in other regions without mishap. Rather it is mentioned here as being one of a number of stresses which are imposed on the animals between the time of being caught and arrival at their final destination.

#### 5.2.2. Blood protein levels during food deprivation

Crab blood, known as haemolymph, may contain a number of different proteins. The most important of these, the pigment haemocyanin, can form from 75-100% of blood proteins. Its main function is that of a transport medium carrying oxygen around the body in a similar way to haemoglobin in the mammalian system. These blood protein levels are highly variable - even among apparently identical animals caught in the same string of pots. This variability gives a clue to the other main function of the pigment - that of being a protein food reserve (see Uglow, 1969).

In the laboratory, the rate and extent of blood protein Loss due to food deprivation at 160C was assessed over a 20-day period. The data are given in Table II and reveal that both the total protein and the haemocyanin are slightly decreased after 1 week and are substantially less after 20 days - the total protein by 22% and the haemocyanin by some 26%. This provides some evidence that the animal is starting to use its blood protein reserves as a food source as a response to starvation. This process, known as catabolism, can also apply to the hepatopancreas - the brown meat - and, eventually, to muscle tissue.

Again, these findings are put forward to demonstrate the extent of the effect of food deprivation on crabs. It should be remembered that these figures refer to crabs which, apart from the absence of food, were being kept in comparatively good conditions of flowing, fully aerated seawater and at a low stocking density. It seems certain that the stresses associated with shipment would elevate metabolic rates and so increase the rate of blood protein losses.

#### 5.2.3. Some effects of aerial exposure

After capture, there are periods of time when crabs are held out of water - in humid conditions. An earlier report (SFIA Technical Report, No. 259) identified aerial exposure as a major source of stress and ultimately the mortality of velvet crabs. The same report provided some estimate of mortality of brown crabs during aerial exposure. Although brown crabs are much more tolerant of aerial exposure than are velvets, such treatments, nevertheless, are major stresses to *Cancer* and should be avoided wherever possible.

The difficulty that aerial exposure introduces to crustaceans is that, under such conditions, they are unable to effect the exchange of gases via the surface of their gills. They can, therefore, neither take in oxygen from their environment nor excrete certain waste products. Respiration at these very Low oxygen

levels is described as anaerobic respiration and, in *Cancer, l*eads to the buildup of a characteristic waste product whose concentration in the haemolymph can be measured. This substance, Lactate, is also toxic to the crab above certain Levels. Experiments were therefore carried out to determine the effects of aerial exposure on oxygen consumption, blood lactate and blood glucose levels for brown and velvet crabs.

The data on oxygen consumption are given in Table III which shows that both species suffer Large decreases in humid air, compared with values obtained in seawater of the same temperature. All measurements were made some 3 days after feeding the animals and refer to quiescent animals. In the aquatic experiments, the animals were allowed 18 hours to settle-down in the respirometers before any recordings were made.

The drop in oxygen consumption in air is accompanied by a build-up of lactate and glucose in the blood of both species (see Table IV). The increases are clear after only 4 hours of exposure and are still evident, particularly in velvets, after 4 hours of re-immersion in fully-aerated seawater. The drop in blood lactate levels which occurs following re-immersion in seawater signifies the repayment of an oxygen debt incurred whilst in air when the oxygen the animals were able to obtain was insufficient. The glucose levels reaching high values when in air are a clear indication that the animals are being stressed at such times.

Normally it would be the case that brown crabs would be subjected to periods of aerial exposure some considerable time after they were captured - hence since they Last fed. The effects of aerial exposure on 3-day, and 10-day starved animals were, therefore, compared in terms of their blood Lactate and glucose levels. The results obtained are given in Table V and suggest that the 10-day starved animals are the more affected. This indicates that these two adverse holding conditions can be mutually reinforcing in terms of the risk which they present.

#### 5.2.4. General considerations of live crab transport

The physical and chemical characteristics of the water used for holding and shipping brown crabs can markedly affect their physiology, hence condition and ultimate survival. Preliminary observations had shown that the salinity of the seawater for holding and shipping the crabs used at the Scottish, Welsh and Spanish premises was in the range 30-34%,s and was unlikely to be a principal cause for concern. Temperature, however, is much more variable and some temperature-dependent affects on brown crab physiology were investigated.

#### 5.2.5. Temperature dependence of oxygen uptake (MO<sub>2</sub>)

The capacity of full salinity seawater to carry oxygen is highly temperature dependent as is shown in Figure 1; at higher temperatures water can carry significantly less oxygen. Conversely the oxygen uptake by crabs increases with temperature because their metabolic rate is temperature-dependent. From the data in Table VI and Figure 2, it can be seen that active or recently-stressed animals have significantly higher oxygen uptake levels than animals which have been allowed to settle down for several hours. It is interesting to

speculate whether the stresses of high packing densities, noise and vibration encountered during transport would ever allow the animals to become unstressed - it seems unlikely.

The data in Table VI also show that raising the holding temperature from 8°C to 12°C increases oxygen consumption by 30-50%. Raising the temperature from 8°C to 16°C roughly doubles oxygen consumption. These last two temperatures correspond roughly with those in the tanks of vivier lorries and the Spanish, land-based holding tanks.

These findings strongly reinforce the importance of maintaining low temperatures during shipment with 8°C being a good target temperature. The apparent gap between the oxygen consumption values of stressed and settled crab is markedly less at low temperatures and reflects the temperature-dependent narrowing of the animals' scope for metabolic activity as a response to stress.

#### 5.2.6. Temperature dependence of ammonia excretion

Ammonia is a by-product of protein metabolism and is produced by crabs. It becomes toxic above certain levels although normally, in the sea, such concentrations are very unlikely to ever occur. However, in the more Limited volumes of water used during holding and shipment, toxic concentrations are likely to occur regularly. Ammonia, however, is a very mobile substance and is readily Lost to the atmosphere by diffusion - particularly if the aeration system is vigorous. Thus, in the absence of filters which would remove it, ammonia concentrations in vivier tanks are dependent on a combination of the rates of production by the crabs and the rates of diffusion to the atmosphere:

Ammonia Concentration = (Rate of Ammonia \_ Rate of Ammonia )

in tank (Production by crabs Diffusion to atmos)

Each component of this equation is variable. The ammonia production by the crabs is dependent upon the water temperature and the number of crabs per unit voume of water. Similarly, the diffusion loss is dependent upon the rate of production and the vigour of the aeration system.

To investigate the possible effects of ammonia build-up, a number of simulations were made using crab to water volume ratios similar to those used during vivier shipment. These experiments showed that sufficiently vigorous aeration could almost compensate for the ammonia production. The build-up of ammonia concentrations was found to be slow, though slightly higher at 14°C than at 9.5°C as shown in Table VIIa.

Simulations were also made by including recently killed animals in the tank when it was found that the build-up of ammonia concentrations was markedly more rapid - see Table VIIb. This simulation was based on a transport mortality level of 1 crab for every 5 litres of water which approximates to 240 dead animals in a loaded tank of 2m<sup>3</sup> capacity and a water:animal ratio of 2:1.

Again it is worth noting that both the ammonia increase, and the concomittant drop in pH, were markedly greater at 14°C than at 9.5° C.This is a point to consider during periods when animals are being held under non-cooled

conditions - for example whilst awaiting packing on vivier lorries or being held by importers.

### 5.3. Description of a Typical Chain of Events from First Sale in Scotland to arrival in Santander

On the occasion of this visit to Benbecula crabs were being purchased from fishermen at various places on the island on a Saturday morning. The crabs were packed dry in plastic containers and remained thus for perhaps 6-7 hours before purchases were brought to the exporter's holding tanks. Animals were stored overnight in the holding tanks which were supplied with running aerated seawater. All the weekend's purchases were packed during the early hours of Monday morning and, on this occasion, were sent dry to Holyhead where the normal arrival would be around 7-8 o'clock on Tuesday morning. This company would also normally expect to receive brown crabs from Scotland that had travelled in tanks in their vivier lorry. These animals would be sorted and packed onto a Spanish vivier for onward transmission to Spain where they arrive at around midday the following Thursday.

Blood samples from randomly selected crabs were taken on the following occasions:

- a) At Benbecula when the Saturday's purchases arrived at the buyer's premises after being out of water for more than 6 hours.
- b) At Benbecula early Monday morning after the Saturday's purchases had been held in the holding tanks.
- c) At Benbecula early Monday morning from crabs brought from Barra the same morning under dry conditions.
- d) At Holyhead from animals brought dry from Benbecula and, a fortnight Later, of crabs brought from Benbecula by vivier Lorry.
- e) On the quayside at Santander as the lorries arrived in Spain. (having been held in vivier conditions all the way from Benbecula).

The blood samples were centrifuged and assayed for total protein, haemocyanin and blood lactate.

#### Blood protein and haemocyanin

Both the total protein and the haemocyanin concentrations were found to be extremely variable. Of the animals bled at Benbecula, total protein concentrations varied between 10.7 and109.7 mg.ml<sup>-1</sup> and haemocyanin concentrations between 10.7 and 85.8 mg.ml<sup>-1</sup>. This large variability existed even though there was no noticeable difference in appearance between the randomly selected animals. The data collected on protein and haemmocyanin values at the various times are summarised in Table VIII. These data show no evidence of any drop in protein levels occurring between Scotland and Wales but it may be significant that the lowest mean value obtained was for the Santander sample, although more samples would need to be taken to confirm this suggestion.

#### Blood lactate values

The data obtained on blood lactate values for the various samples are given in Table IX. These show that the animals which had experienced aerial exposure

when purchased at Benbecula were stressed because recovery values after being held in the tanks are much lower than those taken at the time the animals were received at premises. There is same evidence too that the Barra consignment was stressed on arrival at Benbecula. The Holyhead samples show clearly the advantages of using a refrigerated vivier system; the 'dry' consignment had very high lactate values compared with those of the 'vivier' consignment which was similar to the value of 'recovered' animals at Benbecula. The lactate values at Santander are also very law and provide little indication of any hypoxic stress occurring with animals sent by this method. The animals sampled at Santander were very lively and apparently in excellent condition. It is difficult to envisage how such animals would show rapid deterioration of condition and apparently, this consignment did not show a high rate of mortality.

#### 6. Discussion

A number of factors have been identified so far in this report as causing some degree of stress response in crabs. They have all been identified within the distribution system between the UK and its export markets and, for clarity, they are summarised here.

Location/Event	Factor	Result
Handling at sea Selection and storage at sea	Mechanical damage Mechanical damager	Bleeding/infection Bleeding/infection
First holding:store pots	Salinity variations Sediments Overpacking Starvation <sup>1</sup>	Osmotic embarrassment Clogged gills Mild hypoxia Catabolism <sup>2</sup>
Landing and first transport	Increased ambient temperature Air exposure	Increased metabolic rate, anaerobic resp. Dehydration and gill damage
Selection and grading ashore and repacking	Mechanical damage Changes in temperature Air exposure	Bleeding/infection Changes in met. Rate Anaerobic respiration
Export transport Deaths in transit	Low dissolved oxygen level, high ammonia level, water stagnation, bacterial growth	Anaerobic respiration Toxaemia Infection
Reception at destination	Increase in ambient temp. Other water changes? Handling	Increased metabolic rate possibly leading to catabolism
Season and Sex	Depleted body food reserves Poor feeding condition and availability of food	Low blood protein levels and hepato- pancreas weight, animal in vulnerable condition

Several points emerge. from this analysis, there are three types of factor identified human handling, equipment and crab condition - each of which is amenable to a different degree of correction.

The first degree of correction is the human handling factor. As noted in Section 4 handling is not particularly bad but what room for improvement there is can be achieved through some sort of training. A video on good handling practices is being produced, a series of seminars is being organised, and the Open Tech module on live shellfish transport should go some way towards an upgrading of standards. There is, of course, a limit on the amount of care that can be taken but it has been observed that mechanical damage can be inflicted almost unwittingly. Small cracks in the carapace or damage to the tips of the walking legs can result in heavy blood loss. Crab should be checked at each opportunity and suspect animals rejected. It is, perhaps, worth noting that up to 30% of a crab's weight can be made up of haemolymph and that coagulation (clotting) often does not occur at the site of wounds. Similarly, nicking is bound to weaken animals to be no alternative to it.

Aerial exposure must always be minimised, especially for velvet crabs, but generally it is not thought that it is occuring excessively for brown crab. During these studies there were occasions when periods of exposure were too long but it seems unlikely to be a dominant cause of the types of mortality that are reported on the continent. A

<sup>&</sup>lt;sup>1</sup> Starvation as a stress factor will continue throughout

<sup>&</sup>lt;sup>2</sup> Catabolism is use of body tissues as a food source

number of options are open to minimise the periods of exposure and some of these are discussed below.

Food deprivation is likewise known to stress the animals but is an inevitable result of the current somewhat chaotic variations in supply and demand. It seems likely that the improvements which are coming will regularise these variations and result in an overall reduction in storage times prior to first sale.

At the same stage, the choice of a site for the store pots needs to be made carefully. There are no good reasons for a wrong choice being made given that turbidity, bottom features and salinity can all be checked fairly easily. If an awareness of the problem exists then the combined expertise of Fishermen's Organisations, Development Officers and fishermen themselves must be adequate to counter it.

The second major category of problem is in the handling and holding equipment used. Again it should be noted here that when all other arrangements go well, the current level of equipment is adequate. It is when the ambient conditions are unfavourable and arrangements go wrong that crab may be subjected to particularly stressful conditions. The difficulties generally occur at points of transfer suggesting the need for the provision of either intermediate holding facilities in water or the development of a portable handling unit in which the animals can be maintained in good conditions throughout their journey.

There is no evidence that an upgrading of all of the different elements of the system would prove cost effective but rather that attention to monitoring of conditions in the system will provide the necessary evidence of problems and priorities for change. It is possible that rearrangement of the logistics of the current exercise with concentration of activities at a Limited number of well designed and monitored holding facilities will prove most beneficial. The SFIA and other development agencies could play a role here.

The underlying principle in all this is that the UK part of the chain should maintain control of the commodity and the conditions under which it is held for as Long as possible. Given that the mortality problem arises from a number of factors it is important to reduce the uncertainty within the distribution chain to a minimum. One strategem to effect this would be to promote a British capability in those skills necessary to design and manufacture the various components of the transport system. This is made an even more attractive proposition by the increasing transport and trade in chilled fin fish within the European Community. Many ideas in truck design are now available which enable a very flexible system to be built allowing for varying proportions of load components. This introduces the opportunity of specialist transport for return loads as a means of offsetting export costs. The only disincentive to developments of this type is the current lack of harmonisation of axle loads for some classes of UK lorries relative to those obtaining in Continental Europe. The higher axle loads permitted to those operators mean higher payloads, the profit on which can offset a considerable proportion of the transport costs that would have to be borne by a UK operator. One way of reducing this disadvantage to UK exporters is the development of a cascade system as referred to in section 8 (iv). This would result in a considerable reduction in the proportion of water carried, hence an increase in the useful payload.

The third main category is far more difficult to resolve - indeed currently it is virtually impossible. The problem area seems to be centred on how seasonal variations in physiological condition may predispose some crab to premature mortality. More

simply: are crab from some areas more vulnerable to stress at certain times of the year than others? The difficulty in giving a straightforward 'yes' or no, answer is based on the fact that crabs - and shellfish in general - are neglected species as far as a comprehensive basic biological data-base is concerned. A limited amount of work has been done on some market-orientated characteristics of crab from some areas : sex ratios, meat yields, etc. Nothing, however, exists to describe the more basic biological reasons for the varions in seasonality of breeding cycle, body reserves and the like which mayb e based on 'real' physical differences between crabs of the same species from different areas. Differences which, in this context, might be sufficient to designate crab from some areas as sub-stocks or even sub-species of the main European stock.

In addition to this the live trade in crab from the Highlands and Islands Region has not been in existence Long enough for any pattern to emerge on the seasonality of mortalities. Thus, when a period of high mortalities starts, no reassurance can be given to the purchaser about how long- or short-lived it might be.

In an attempt to ease some of these uncertainties proposals have recently been made to initiate a long-term comparative study of the variations in condition of brown crab from several areas. An examination is also to be made of the feasibility of devising a simple method of condition assessment. This might, for example, take the form of some sort of conductivity test or the use of a test reagent in holding water. Whatever might transpire, the attractions of such a test are obvious. It would enable an assessment to be made of the animals' chances of surviving the export process.

One of the ironies of the current situation in the W. Isles is that it is likely that the individuals most at risk are those that have the highest value. These are the hen crabs that are just ready to spawn and have a very high proportion of coral - the egg mass - in the carapace. As noted earlier these are the crabs that are judged to be in good condition or quality as far as their value whilst alive is concerned, but in very bad condition in respect of their resistance to stress.

The only other area of uncertainty is that of reception conditions in Spain. During the visit to Northern Spain in 1985 it was only possible to make a very brief check on conditions at one location. Perfectly reasonable assumptions can, however, be made about the extent of thermal shock that must occur at this stage in the process. It is known for example that special chilled reception tanks were built for velvet crabs because they died when taken from cooled vivier tanks and put straight into Spanish seawater tanks at ambient temperature. It is easy to surmise that the slightly less sensitive brown crab takes longer to die but suffers the same eventual fate.

In these circumstances there is a strong case to be made out for either an SFIA staffed examination of this aspect of the system or, possibly, a cooperative effort between SFIA and the Spanish Oceanographic Institute.

#### 7. Conclusions

A number of possible causes were identified for the excessive mortalities of brown crab during transport to their continental destinations. Of these no single factor could be blamed, rather it was concluded that three or four contributed to these mortality levels. The following were strongly implicated:

- i) some physiological or condition factor which, at certain times of the year, results in crabs being unable to withstand high stress levels. The most likely effect of this seasonal influence is that the crab have a very low level of body reserves to call on when stress requires them to be alert and active.
- ii) difficulties experienced during first holding and/or transfer between holding/transport phases. These are most likely to involve low salinity conditions or excessive exposure to air.
- iii) general weakening of the animals through a combination of blood loss from 'nicking' and mild starvation. The procedures which result in this are currently unavoidable.
- iv) the presence of dead animals in holding tanks, particularly those on lorries, causing a rapid build-up of ammonia products in the holding water.
- v) the stress imposed by transfer to Spanish holding tanks where the water temperature is significantly higher than that of the lorry tanks.
- vi) a number of alternative transport regimes were looked at including air, rail and hybrid systems. None offered the advantages and flexibility of a well-designed road freight system.

It was noted that the monitoring device developed by the SFIA proved useful in identifying problems in holding and transportation that would otherwise not have been located. The use of such devices, in both shore and vehicle installations, is seen as a necessary part of adequate control of the distribution system.

#### 8. Recommended Further Action

- i) an overall, long term aim of any development should be to achieve the maximum UK-owned control over the distribution process. This would minimise the current uncertainty over transport conditions and mortality rates and should present opportunities for profitable return loads.
- ii) the handling, holding and transport system needs upgrading if only to cope better with various contingencies. First examinations have shown a fully unitised system to be impracticable. The most useful improvements would be the building of more centralised intermediate holding tanks and the part mechanisation of the loading/offloading procedures. These two actions would be both cheap and effective ways of reducing handling and air exposure.
- iii) the development of a UK capability in the building of vivier handling systems would be advantageous. Not only is the market for Live crab increasing, but demand is growing for other species too.
- iv) a further study should look at ways of increasing the Loading factor on the vivier Lorries. A 'cascade system' of stacked trays with water running down through them seems to offer considerable advantages. These include making full use of the height of the truck body and reducing water from 50% to less than 20% of the load.
- v) more biological/stock investigations are needed if rational development policies are to be devised. It is difficult to even state a good cost- benefit case when much basic information is simply not available. In the first instance a study of all the various exploited stocks needs to be undertaken in respect of those basic characteristics which influence their marketability.







Fig. 2 – Oxygen Uptake by *Cancer pagurus* at different holding temperatures in 'Active' and 'Settled' states

## Table 1Cancer pagurus: Oxygen uptake of 'settled' animals at 2 and 10 days post-feeding. Values are means ± Standard Errors for replicate tests on 4 animals

Temp.	Settled MO <sub>2</sub> (µI 0 <sub>2</sub> .g <sup>-1</sup> .hr <sup>-1</sup> )		
(°C)	3 days post-feeding	10 days post-feeding	
16	41.8 ± 1.81	34.31 ± 9.12	

# Table 11Cancer pagurus: Changes in blood protein and haemocyanin concentrations<br/>during starvation. Values are given as means ± Standard Errors for n = 9<br/>animals in each case

Day since being fed	Total protein (mg.ml <sup>.1</sup> )	Haemocyanin (mg.ml <sup>.1</sup> )				
0	83.86 ± 6.88	77.66 ± 7.02				
7	79.32 ± 6.22	76.54 ± 6.67				
20	64.99 ± 4.94	57.53 ± 4.98				

#### Table III

#### Oxygen consumption values for brown and velvet crabs in water and in air. Values are means $\pm$ Standard Errors of 5 animals in each case. (T = 16°C, S = 35% and relative humidity – 75-80%)

Spacios	MO <sub>2</sub> (µI 0 <sub>2</sub> g <sup>-1</sup> .hr <sup>-1</sup> )		
Species	In water	In air	
C. pagurus	41.83 ± 1.81 (100%)	5.69 ± 0.32 (13.6%)	
N. puber	48.66 ± 8.58 (100%)	9.81 ± 0.96 (20.2%)	

# Table IVCancer pagurus and Necora puber: Blood lactate and glucose concentrations in mg. $100 \text{mm}^{-1}$ during normoxia and during aerial exposure and reimmersion. Vlaues are given as means ±<br/>Standard Errors for 6 animals in each case. (T = $16^{\circ}$ C; Relative Humidity = 75-80%)

	Timo	C. pa	gurus	N. p	ouber	
	(hours)	Normoxia	Aerial Exposure and Reimmersion	Normoxia	Aerial Exposure and Reimmersion	
	0	5.58 ± 0.46	10.34 ± 0.41	4.57 ± 0.35	4.86 ± 0.55	
Lactato	4	6.67 ± 0.34	34.84 ± 3.56	4.51 ± 0.16	58.97 ± 3.22	Doimmorsion
Laciale	8	7.29 ± 0.49	14.73 ± 7.41	4.22 ± 0.13	26.03 ± 3.70	Keimmersion
	10	-	-	-	16.30 ± 6.50	
	0	3.24 ± 0.67	10.14 ± 1.17	3.780 ± 1.24	1.56 ± 0.23	
Clucoso	4	1.83 ± 0.33	24.35 ± 3.02	1.95 ± 0.32	11.60 ± 2.59	Doimmorsion
Glucose	8	5.76 ± 2.08	17.22 ± 2.07	$2.06 \pm 0.20$	9.60 ± 2.21	Reminersion
	10	-	-	-	8.26 ± 3.49	

#### Table V

## *Cancer pagurus:* The effects of aerial exposure and reimmersion on the blood lactate and glucose levels (in mg. 100ml -1) of animals with different recent feeding histories.

#### Values are given as means $\pm$ Standard Errors for n = 6 animals in each case

	Time	3 days post-feeding	10 days post-feeding
Lactate	0 hr. (Normoxia)	10.34 ± 0.41	11.27 ± 1.11
	4 hr. (Aerial Exposure)	34.84 ± 3.56	39.69 ± 2.95
	4 hr. (Aerial Reimmersion)	14.73 ± 7.41	14.09 ± 5.49
Glucose	0 hr. (Normoxia)	10.14 ± 1.17	15.45 ± 1.25
	4 hr. (Aerial Expsoure)	24.35 ± 3.02	27.24 ± 2.21
	4 hr. (After Reimmersion)	17.22 ± 2.07	19.24 ± 1.80

#### Table VI

### *Cancer pagurus:* Oxygen consumption values of 'active' (1 hr post-handling) and 'settled' (6 hr post-handling) animals at 3 seawater temperatures. Values are given as means ± Standard Errors of replicate tests on 3 animals on each case

Temp (°C)	'Active' animals	MO <sub>2</sub> (µl 0 <sub>2</sub> g <sup>-1</sup> .hr <sup>-1</sup> ) 'Settled' animals	Difference (%)
8.0	23.94 ± 0.63	3.00	65.0
12.0	5.92	0.65	57.9
16.0	4.42	9.12	77.4

Table VIIACancer pagurus: Ammonia production by 15 dead animals in a tank containing 26.5 litres of seawater (3.5 litres<br/>displacement). The animal to water ratio = 1:1.8

Time (hr)	Temp (°C)	0₂Level (%)	Salinity (%, S)	рН	Ammonia Concentration (mg. 1-1)	mV
0	10.0	81.4	35.2	7.112	0.136 (8.0 x 10 ⁻⁰M)	97.9
4	9.5	78.7	-	6.910	-	-
24	9.5	78.2	35.2	7.105	0.153 (9.0 x 10 <sup>-6</sup> M)	94.9
				Change = 0.017		
0	14.0	89.8	34.2	7.609	0.133 (7.8 x 10 <sup>-6</sup> M)	99.1
4	14.0	90.5	-	7.048	-	-
24	14.0	54.0	34.5	7.235	0.162 (9.5 x 10 <sup>-6</sup> M)	93.7
				Change = 0.029		

## Table VIIbCancer pagurus: Ammonia release by 1 dead animal in a tank containing 5litres of seawater and over a 24 hr. period

Time (hr)	Temp. (°C)	рН	Ammonia Concentration (mg.1-1)	mV
0	9.5	7.378	0.136 (0.8 x 10⁻⁵M)	94.8
24	9.5	7.084	0.170 (1.0 x 10 <sup>-5M)</sup>	91.2
		Change =	0.024	
0	14.0	7.050	0.425 (2.5 x 10 ⁻₅M)	84.3
24	14.0	6.450	1.649 (9.7 x 10 <sup>-5</sup> M)	62.3
		Change =	1.224	

#### Table VIII

#### Cancer pagurus: Blood protein and haemocyanin concentrations of samples taken at various points of the transport route between Benbecula and Santander. Values are given as means ± Standard Errors

Sample Details	n	Blood Protein (mg.ml <sup>-1</sup> )	Haemocyanin (Mg.ml <sup>.1</sup> )
At Benbecula	29	43.78 ± 3.81	40.73 ± 3.23
At Benbecula – sample from Barra	20	53.86 ± 6.18	53.38 ± 4.72
Holyhead – Benbecula sample brought 'dry'	18	47.96 ± 3.89	41.87 ± 3.68
Holyhead – Benbecula sample brought by vivier	10	59.08 ± 6.16	45.16 ± 6.41
Santander	8	40.10 ± 6.46	29.30 ± 4.74

#### Table IX

# Cancer pagurus: Blood lactate concentrations of samples taken at various points of the transport route between Benbecula and Santander. Values are given as means ± Standard Errors. Numbers in parentheses refer to the number of animals sampled.

Sample Details	Blood Lactate Concentration (mb. 100ml-1)
Benbecula – after more than 6 hours aerial exposure	40.24 ±3.04 (19)
Same batch of animals as above – after more than 30 hours in holding tanks	17.15 ± 2.88 (9)
Benbecula – animals brought from Barra by vivier	31.95 ± 2.88 (20)
Holyhead – sample brought 'dry' from Benbecula	63.62 ± 3.21 (24)
Holyhead – sample brought by vivier from Benbecula	20.42 ± 2.29 (10)
Santander	11.75 ± 1.96 (8)

#### Appendix I

Description of the Electronic Data Acquisition System

#### **Description Of The Electronic Data Acquisition System**

To enable continuous examination of water quality during the various stages of the transport system for live shellfish a four-channel data acquisition system has been constructed in the SFIA electronic laboratory.

The four analogue input channels will accept variable voltage inputs from temperature sensors, pH and dissolved oxygen probes within the range  $\pm$  25 volts. Other types of sensor can be accepted as necessary. Each channel is able to store 1000 readings, scanning rates are selectable from 1 second to 15 minutes in 1 second steps. Logged data can be transferred to tape, chart recorder or microcomputer for analysis.

Power for the microprocessor controller and interface unit is derived from external batteries with capacity for 48 hours continuous recording. Should there be a loss of power then data already logged will be saved in memory for not less than 72 hours by the internal battery back-up.

The first field test of the system was aboard a vivier truck between Islay and Holyhead. It was set up to monitor the temperature and dissolved oxygen in two tanks containing velvet crabs. The system was programmed to take a set of readings every two minutes. A print out from this journey is shown as figure A1.1 and some interesting points emerge from it.

It was difficult at first to explain the series of sudden drops in the oxygen saturation levels although the driver had found it necessary to tighten the belt to the compressor a few days after this trip. It was eventually established that there had been heavy rain during the periods in question and, although the belt was not slipping when dry, the rain was enough to seriously affect the operation of the compressor.

Presumably, had there been heavy rain throughout the journey, the cargo would have been in very real jeopardy. This state of affairs might then have gone completely unnoticed by those concerned. The advantage of a data display and logging system such as this is that it can immediately alert the driver to this sort of situation and can also be used as evidence that water quality has been maintained throughout the journey.



Fig. A1.1 Dissolved oxygen trace made during journey by vivier truck

Appendix II Illustrations of Holding Facilities



\* Damage to Legs Can Be 'Repaired' by Casting; Damage Inside the Casting Point Cannot Be Made Good







Fig. All.1 - Live holding in store pot and deck well





Fig. All.2 – Spanish vivier lorry





Fig. All.3 – Crab and lobster holding tanks



Fig. All.4 - Dead crab examined for damage to limbs