

Farming soles – a reality at last?

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Introduction

The farming of soles may at last be on the verge of commercial viability, but the third in a series of workshops held at of CIFPA El Toruño, Cadiz, Spain earlier this year highlighted some of the obstacles yet to be overcome before this can become a reality.

The farming of soles has been an aspiration of aquaculturists in Europe since Shelbourne's pioneering work at Port Erin, Isle of Man almost half a century ago. Both the common (*Solea solea*) and the Senegal (*S. senegalensis*) sole are attractive species because of their high market value and relative ease with which juveniles can be mass-reared in hatcheries. Sadly, they have proved rather less amenable during subsequent on-growing stages, poor growth and survival rates being largely attributable to the problems of nourishing these choosy feeders. For this reason, commercial success has been limited to species, such as sea bass, sea bream and turbot, all of which offer fewer on-growing problems than soles.



Solea senegalensis broodstock at CIFPA El Toruño, Cadiz, Spain
(from Bari Howell)

The situation changed during the late 1990s. The problems of feeding juvenile soles on formulated feeds were largely removed by developments of specialist feeds produced by novel methodologies. This coincided with a commercial imperative of the bass/bream industry to diversify into other species to combat market saturation of those species. Interest in farming soles increased and in 2002 a first workshop was organised to re-appraise the prospects for commercial farming of these species.

Sole workshops I and II

This first workshop was held at the CEFAS laboratory in Weymouth, UK where an 'expert' group of invited practitioners from the commercial and research sectors critically examined each phase of production and identified areas of weakness that required further research and development. The general conclusions were that:

- Techniques for culturing sole over the last ten years or so have made significant advances and most of the major constraints (mainly related to the provision of commercially acceptable formulated feeds) have been removed. This has been reflected in a much greater involvement of commercial companies than ever before.
- *S. senegalensis* displays higher growth rates and apparently higher tolerance to intensive culture conditions than *S. solea* and is consequently much better suited to commercialisation at this time.
- *S. solea* will only be commercially farmed if certain biological problems (growth rate and fin erosion) in culture are solved, but only then if sufficient marketing

opportunities are found in which this species has a distinct commercial advantage over *S. senegalensis*.

The workshop generated considerable optimism in the prospects for developing commercially viable systems. Over the next year or so, however, serious obstacles to reaching that goal emerged. In particular, widespread spawning failure of captive stocks limited the availability of eggs and larvae to support both commercial trials and experimental programmes. Furthermore, at sites where juveniles were available for growth trials, disease proved to be a major cause of mortalities which in many cases decimated stocks. As a result of these experiences a second workshop was organised in September 2003 to allow a detailed examination of the specific issues of reproduction and disease. On this occasion the meeting was held at CIFPA (CICEM at that time) El Toruño, Cadiz, one of the few sites at which regular spawning of sole was being obtained. The main discussions were summarised as follows:



Dr Jose Pedro Cañavate describing the facilities at CIFPA El Toruño, Cadiz, Spain during the second sole workshop in 2003 (from Bari Howell)

- For *Solea senegalensis*, an analysis of egg production data in industry and research facilities suggested the importance of winter temperatures as a determinant of successful fertilised egg production. The best egg production was obtained when winter temperatures had fallen to 10-11°C and had remained below 13°C for six weeks. Complimentary research programmes provided evidence that this factor may be of particular importance for successful maturation of males. High stocking density was also identified as a possible important cause of egg production failure. The use of reared fish and formulated feeds were highlighted as novel aspects of broodstock management whose efficacy is yet to be demonstrated, though no unequivocal evidence of their unsuitability was evident. These factors also appear to be key determinants of fertilised egg production in *S. solea*.
- Pasteurellosis, vibriosis and myxobacteriosis are emerging as the most important bacterial diseases of soles. These species are also susceptible to nodavirus and birnavirus viral pathogens. Reduction of stress in culture systems was acknowledged to be an important preventative measure and in this context high stocking density was identified as one of the most important stressors. In flatfish, however, it was considered that communal conditions may to some extent relieve stress associated with the absence of sand, and that the damaging effects of high density may be due as much to deterioration in water quality as to social interactions. There is an urgent need for the identification and development of suitable vaccines and other prophylactic measures for protection against the important pathogens of soles.

In the two years following this second workshop significant improvement in the problem of egg supply was reported, but disease continued to be a major problem at almost all production sites with major fish losses threatening the commercialisation of these potentially important species. These problems in particular generated the momentum for a third workshop during March 2006.

Sole workshop III

This Workshop was again held at the facilities of CIFPA El Toruño in Cadiz and was attended by 46 participants from both the research (59%) and commercial (41%) communities. Not surprisingly, Spanish participants outnumbered the rest by about 2 to 1, but five other European countries were represented (Greece, Holland, Norway, Portugal and the UK) indicating the widespread interest in these species. With this mix of nationality and background the Workshop represented a good cross-section of European interests in the culture of these species. Bari Howell again organised and chaired the Workshop, ably supported by the co-authors of this report.

The purpose of the meeting was *to review the current status of commercial experience and corresponding research in key areas of the cultivation of soles and to provide information that would guide the future actions of both sectors.*

The primary area of concern was disease and the whole of the first day of the two-day meeting was dedicated to that subject. The second day was dedicated to a review of reproduction as well as presentations on other important areas of science that will help to underpin the future of this developing industry. The workshop closed after a wash-up session during which the participants from industry presented their views on research priorities and the rearing requirements of both *S. senegalensis* and *S. solea*.

Discussions were informed by presentations, the abstracts of most of which are included in the full report of the workshop (on the EAS site at www.easonline.org)



Some of the participants at the third sole workshop multi-tasking in the bar of the hotel (from Bari Howell).

Disease

The session on disease comprised presentations on both diagnosis and prevalence of disease as well as on prevention and control. The main conclusions agreed by participants were as follows:

- Reports of opportunistic diseases confirmed that a number of diseases remain prevalent and are a significant threat to progress towards the commercialisation of both *Solea senegalensis* and *S.*



Left: Symptoms of black patch necrosis in *Solea senegalensis* (from Carlos Zarza)

Below: Reared *Solea solea* in experimental facilities in the UK showing badly eroded tails probably as a result of *Flexibacter* sp infection (from Stephen Baynes).

solea. Diseases of particular significance include pasteurellosis, vibriosis and flexibacteriosis.

- Pasteurellosis, a particularly intransigent condition, tends to be restricted to Mediterranean locations where water temperatures are higher and proximity to susceptible species such as sea bream is more likely. In locations on the Atlantic coast (i.e. lower temperature) diseases caused by *Tenacibaculum* (Flexibacteriosis) were more prominent.
- Although viral diseases have not yet achieved the prominence of bacterial diseases, Viral Nervous Necrosis (Nodavirus) has been detected in soles in the UK, Italy and possibly Spain. Other viruses have been isolated from *S. senegalensis* and characterised.
- In contrast to most farming locations, these diseases were reported to be relatively unimportant in a recirculation facility housing *S. solea* (one site). In this situation other infestations, such as ciliates and dinoflagellates, linked to the biofiltration system had caused some relatively minor problems.
- It was clear from other reports that specialist resources at a number of centres of excellence have been mobilised to develop methods for preventing and/or controlling such disease outbreaks. Reports were received on positive results on the use of probiotics and immunomodulators which demonstrated significantly improved survival in controlled challenge tests.
- The potential of vaccines was also emphasised. Vaccines against pasteurellosis, are licensed for sea bass (Greece). There is a need to develop vaccination protocols and vaccination regimes that fit with disease dynamics in sole. By optimal vaccination protocols, a significant reduction of mortality should be obtained, helping the industry becoming more sustainable. It may however, be difficult to eradicate the pasteurellosis from sole production.
- It was also felt that there was considerable scope for improvement through better husbandry techniques. In particular, greater consideration should be given to the specific requirements of the species arising from its



nocturnal behaviour, mode of feeding and environmental requirements. The need to improve juvenile quality, as well as the quality of eggs on which hatchery operations depended, was also emphasised. The longer term effects of rearing conditions were recognised. In this context it was felt that extensive pond-rearing could provide a valuable method of benchmarking intensive rearing methods.

- It was considered important that systems should be developed in parallel with husbandry methods that take full account of the characteristics of the target species
- With regard to future commercial developments, strong interest was expressed in the use of recirculation systems which allowed better control of key environmental conditions (e.g. temperature) and bio-security than open flow systems. However, the view was expressed that such systems are not yet fully reliable and potential problems may not have already been appreciated. Alternatively, the use of ground water when available was also considered to be desirable.
- Finally, the value of sharing information of both a positive and negative nature in promoting the development of the industry was emphasised.

Reproduction

Conclusions regarding conditions required for successful spawning are based on both an analysis of spawning records from research and commercial sites and on research projects on specific aspects of reproduction. The majority of the



Shallow raceways being used to grow *Solea solea* in a recirculation on-growing system in Holland (from Andries Kamstra).

information presented was for *S. senegalensis*, reflecting the greater interest in this species, though data for *S. solea* was made available for two sites. The overall conclusions were as follows:

- Within the last three years, natural reproduction of wild caught Senegal sole breeders has extended from a small number of facilities to several other facilities around Spain and Portugal. Egg production is now reported to occur in at least nine facilities dedicated to both research and production.
- The application of controlled water temperature fluctuations were described as the main triggering factor to successfully obtain viable spawning from *Solea senegalensis* held in captivity. However, water temperature stabilisation at 18°C after pre-conditioning at lower or higher temperatures, was used in one facility as a procedure to regulate the timing for spawning.
- Despite the generalised success in spawning of *S. senegalensis*, important differences in fecundity and fertilization still occur between hatcheries. Noticeably, different husbandry conditions together with different origin and physiological status of breeders are likely to be responsible for such differences.
- Previously, egg production from first generation (F1) reared fish had not been reported, but importantly on this occasion, there was one report of naturally produced fertilized eggs from this type of fish. In this instance, similar fertilization rates but lower fecundity and spawning frequency than wild breeders were observed. The use of formulated feeds with this stock also importantly demonstrates that the use of natural feeds for broodstocks is not obligatory.
- In one facility, doubling the male to female ratio to 2:1 did not affect egg fertilization, but increased fecundity. However, in other facility a 1:1 sex ratio was preferred.
- From sexual hormone analyses, it has been proposed that January seems to be a sensitive period for temperature conditioning for subsequent gonadal development of *S. senegalensis*.

- *S. solea* F1 males were found not to complete spermatogenesis, probably due to lower levels of 11kT and a block at the spermatid stage. It was postulated that this might have been caused by the nature of the annual temperature cycle.
- Experiments on hormonal induction of spawning were focussed on F1 broodstock. Results showed that treatment with Gonadotrophin-releasing hormone analogue (GnRH_a), mainly administered via slow-release delivery systems (implants and microspheres), highly stimulated gonad maturation and spawning in both males and females, although no viable eggs (0% hatching) were obtained. On the other hand, fertilized eggs giving rise to viable larvae (F2 generation) were reported to be produced for the first time from F1 breeders, in a broodstock where females and males were treated with GnRH_a implants and human chorionic gonadotrophin (hCG) injections, respectively.
- Strong social interactions within a breeding tank were suggested after microsatellite parentage assignment of larval off-spring throughout a yearly cycle. It was estimated that a 47% reduction of the effective population size occurred in just one generation.

Other research

Although the workshop focussed on the two areas that are currently the major constraints to commercial development, presentations were also received on a wide range of other research topics that are essential to establishing a sound scientific basis that will underpin the development of this industry. These included the role of the pineal and melatonin in synchronising sole reproduction rhythms; feeding, nutrition and quality in *S. senegalensis* post-larvae; growth and size variation, and the application of genomics and proteomics in sole (& halibut) culture.

Industry priorities

With regard to industry's immediate needs, the following research priorities were identified:

- Spawning F1 broodstock & protocols
- Protocols for all year spawning
- 'Bench marking' juvenile quality using pond reared vs hatchery reared fry
- Feed quality
- Management of sole in recirculation systems
- Vaccine development for *Flexibacter* & *Pasteurella*
- Study on development of the immune response in sole
- Studies on the use of probiotics and immuno-modifiers

During the final discussion the participants emphasised the usefulness of this type of forum as a vehicle for exchanging information between researchers and industry and in establishing networks that will facilitate the continuing development of the industry. Serious problems remain, but there was a considerable level of optimism that these are tractable and with an appropriate application of research resources in close collaboration with industry the goal of achieving commercial production of these species will be reached in the near future.