RESUMEN PROYECTO ANIPRESS

El proyecto ANIPRESS explora el potencial del procesado a alta presión como una alternativa a la congelación para la inactivación de larvas de *Anisakis* en productos pesqueros. La presencia de *Anisakis spp*., parásitos nematodos ampliamente distribuidos en ambientes marinos, supone una importante amenaza para los consumidores de pescado crudo o poco cocinado. Tras su ingestión, las larvas penetran en la mucosa gástrica e intestinal, provocando los síntomas de la anisakiasis que incluyen dolor abdominal, vómitos y diarrea. Además, pueden producir respuestas alérgicas en individuos sensibilizados con síntomas que abarcan desde la urticaria y el prurito hasta el shock anafiláctico con peligro vital. Para evitar el riesgo de anisakiasis, la legislación obliga a congelar el pescado que se vaya a consumir crudo o poco cocinado y mantenerlo a una temperatura mínima de −20 °C durante, al menos, 24 h. Sin embargo, la congelación presenta importantes inconvenientes (alto gasto energético, tiempo necesario, degradación en la calidad del producto, entre otros) y, por ello, la Autoridad Europea de Seguridad Alimentaria sugiere la búsqueda de estrategias alternativas que permitan reducir el riesgo de infección por *Anisakis*. En este sentido, el procesado a alta presión ofrece ventajas únicas ya que es mucho más rápido que la congelación, reduce significativamente el consumo de energía y garantiza un tratamiento uniforme en todo el producto.

Para evaluar su viabilidad integrando aspectos tanto de seguridad como de calidad del producto, este proyecto investiga el impacto de las altas presiones no sólo sobre las larvas de *Anisakis* sino también sobre distintos productos pesqueros en distintos escenarios. Así, se desarrollará un modelo cinético capaz de describir la inactivación de las larvas en función de las condiciones de presión/tiempo aplicadas y se estudiarán los cambios en la actividad locomotora, las alteraciones metabólicas y los daños estructurales que sufren las larvas durante el procesado para identificar marcadores que definan su infectividad. Además, el proyecto simulará escenarios del mundo real para evaluar el riesgo de infección en distintos escenarios, incorporando variables como la especie de pescado, la prevalencia de *Anisakis* y la intensidad de infección y determinará, empleando el modelo cinético y los indicadores de infectividad identificados, condiciones de procesado seguras en distintos escenarios. En la etapa final del proyecto, se evaluará la calidad sensorial, microbiológica y fisicoquímica de distintos productos pesqueros tratados con alta presión y se determinará su idoneidad para usos culinarios diversos como consumo en crudo, marinado o cocinado a baja temperatura. Además, los productos presurizados se compararán con sus análogos congelados para, con toda la información recogida, evaluar la viabilidad del procesado a alta presión como alternativa a la congelación.

Con todo ello, ANIPRESS pretende generar conocimiento científico y técnico para ofrecer soluciones dentro de la estrategia "de la granja a la mesa" de la Comisión Europea. El proyecto tiene el potencial de mejorar la seguridad alimentaria en productos pesqueros crudos o poco cocinados, reduciendo el riesgo de anisakiasis y ofreciendo una alternativa más eficiente a la congelación. Esto podría tener un impacto significativo en la industria pesquera y en la salud pública, permitiendo el procesamiento de productos pesqueros crudos de manera más segura y eficiente.

ALGUNAS DE LAS ÚLTIMAS PUBLICACIONES DEL GRUPO RELACIONADAS CON EL TEMA DEL PROYECTO

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Modeling survival curves of *Anisakis* L3 after isothermal heat treatments at lethal temperatures

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ABSTRACT

Even though anisakiasis is considered, nowadays, a significant threat to public health all over the world, no attempt has been made up to date to mathematically describe the thermal susceptibility of *Anisakis* larvae in the third stage (L3). To fill this gap, in this paper, more than 10,000 free (non-encysted) *Anisakis* L3 were individually heat treated in a thermal cycler at temperatures between 44 °C and 61 °C for different exposure times. After heat exposition, viability was assessed in each larva, survival curves at isothermal conditions were derived, and the effectiveness of four kinetic models (fundamental kinetic model, Mafart model, and probit and logit models) in describing these curves was tested. Evaluation of larvae viability after heat exposition revealed sigmoidal survival curves that increased their steepness with temperature. Of the four models tested, the Mafart model was the one that best fitted the data only differing from the observed survival ratios by 0.12 units on average. Validation experiments performed at temperatures different to those used to create the model corroborated its predictive capacity. Future efforts should be focused in predicting larvae viability at non-isothermal conditions as those occurring during fish cooking.

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Behavioral and physiological changes of *Anisakis simplex* complex third stage larvae upon heating

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ABSTRACT

There is increasing information on the time-temperature conditions that can be lethal for *Anisakis* larvae, but little is known about the infective potential of larvae that survive heat treatments. To characterize the risks associated with accidentally undercooked fish, in this paper, more than 5000 *Anisakis* larvae in the third stage (L3) were individually treated, at quasi-isothermal conditions, in a thermal cycler at 50 °C for different exposure times (0–320 s). After heat exposition, surviving larvae were collected and their locomotor activity, resistance to simulated gastric juice (SGJ), agar penetrative ability, and respiration capacity were evaluated. Results show that, even after relatively short treatments that rendered survival ratios ($S_{t_{50^{\circ}C}}$) close to 1 (i.e. all alive), some behavioral and physiological characteristics can be considerably impacted. Thus, L3 locomotor activity and agar penetrative ability significantly decreased after 60 s at 50 °C ($S_{60\ s} = 1$), while heat expositions for 90 s ($S_{90\ s} \approx 0.98$) also reduced the resistance to SGJ and the respiration capacity. Our study reveals that the behavior of heat-treated *Anisakis* larvae depends on the intensity of the treatment and the longer the exposure time at 50 °C, the greater the changes observed in all the tested indicators. Further research is needed to elucidate the implications of the decline of these indicators on infectivity.

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Predicting inactivation of *Anisakis* L3 after non-isothermal heat treatments: Impact of thermal conditions during and after cooking

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ABSTRACT

Accurately predicting the thermal inactivation of *Anisakis* larvae during cooking is essential to ensure food safety while preserving the quality of fishery products. In a previous study, we developed a Weibull-based model that successfully described larval inactivation under isothermal conditions up to 55 °C. However, real cooking processes typically involve dynamic temperature profiles with gradual heating and cooling phases. To estimate *Anisakis* viability under realistic processing conditions, a predictive model capable of integrating the entire non-isothermal thermal history is required.

To do so, in this study, we implemented the differential form of the Weibullian model and explored two alternative formulations to describe how thermal resistance changes with temperature: a single log-quadratic equation and a more flexible piecewise log-quadratic function. All model parameters were optimized using 98 previously published non-isothermal inactivation experiments and validated against 26 independent treatments. While the piecewise model provided a slightly better fit during training, the single log-quadratic formulation showed superior predictive accuracy ($R^2 = 0.9782$, $RMSE = 0.0497$) and was selected for further use.

The validated model was used to investigate how heating and cooling rates affect larval inactivation. Simulations revealed that slow heating and cooling significantly increase lethality, even at mild temperatures. These predictions were consistent with experimental observations and underscore the critical influence of thermal history beyond peak temperature alone. This modeling approach provides a robust framework for designing safe, quality-preserving cooking protocols and offers valuable support for risk assessment and regulatory guidance in seafood safety.