Osteoarthritis (OA), which is characterised by progressive cartilage degradation, is the most common form of arthritis affecting more than 10% of the EU population over 60 years of age. The cause of OA is still not known and there is no cure available. The current treatment involves pain relief and joint replacement surgery.

The age of the population in the EU and worldwide increases continuously, therefore understanding of the molecular processes involved in the pathogenesis and progression of OA is of great importance.

A network of proteins controlled by regulatory proteins is responsible for the proper functioning of living organisms. Ubiquitination is a process where a small protein is attached to other proteins and causes their breakdown or changes their way of acting. Some proteins are known to regulate cartilage production and degradation. WWP2 is a protein which ubiquitinates other proteins, and is known to regulate production of cartilage (chondrogenesis).

We aim to identify and characterise WWP2 interactors and substrates using a combination of proteomics and molecular biology techniques. In addition, we will investigate the role of WWP2 and its substrates in cartilage biology with the goal to uncover molecular targets for OA treatment and other WWP2-related diseases.

A significant amount of time during the reporting period was allocated to generating tools necessary for the project to progress. Using these tools, we identified several potential WWP2 interactors/substrates. The proteins were identified by co-immunoprecipitations (Co-IPs) using the catalytically inactive WWP2 in combination with mass spectrometry. Further, several possible WWP2 substrates were identified in cells depleted of WWP2 (using siRNA) and in cells over-expressing tetracycline-inducible, active WWP2. The proteins were identified using SILAC (stable isotope labelling with amino acids in cell culture) and quantitative mass spectrometry.

Five of the identified proteins were confirmed as WWP2 interactors in co-IPs and/or in-vitro binding assays. In-vitro ubiquitination assays indicate that four of the proteins are ubiquitinated by WWP2.

A subset of the results have been presented as a poster at the 9th Cold Spring Harbor meeting on The Ubiquitin Family, Cold Spring Harbor, New York, USA.

The reported work was performed at the National Institute of Health in the USA and is continued at Newcastle University in the UK. Currently, we are investigating the involvement of the identified proteins in chondrogenesis and chondrocyte biology.