

P026 Localisation of the cytoplasmic sub-domains of the desmosomal cadherin desmoglein 2

Anthea Scothern and David Garrod

Faculty of Life Sciences, University of Manchester

Desmosomes are intercellular junctions found in epithelia and some other tissues. Their primary function is strong cell-cell adhesion. They also link the intermediate filament cytoskeletons between cells and have roles in cell signalling, tissue morphogenesis and wound repair. Their adhesion molecules, the desmosomal cadherins desmoglein (Dsg) and desmocollin, have unique features in their cytoplasmic domains which may be important for desmosomal structure and function.

This project focuses on the cytoplasmic domain of Dsg 2 (Dsg2-Cyt), the largest and most widespread isoform of Dsg. This consists of 483 amino acids and 5 subdomains. The aim of the project is to localize the subdomains within the desmosomal plaque by immunogold-electron microscopy.

Recombinant N-terminally GST-tagged Dsg2 Terminal domain was expressed in *E.coli*, purified and used to raise polyclonal antibodies in rabbits.

These antibodies have been characterised along with a mouse monoclonal antibody, 33-3D. Characterisation has shown that anti-Dsg2TD reacts exclusively with the terminal domain and 33-3D reacts exclusively with the repeat domain of Dsg2. All antibodies recognise the full length Dsg2 molecule found in HaCat (human keratinocyte) cell extracts.

Preliminary quantitative analysis of immunogold labelling of ultrathin cryosections of epidermis indicates that these domains are localised in the outer dense plaque of the desmosome.