Abstract (English)

Oncogenic-induced senescence is considered to be a primary fail-safe mechanism in tumourigenesis of breast cancer. The induction of senescence leads to many structural and metabolic changes as well as cell cycle arrest. Here, we investigated underlying changes that occur as proliferating cells enter senescence through upregulation of the receptor tyrosine kinase ErbB2. To study these changes, NeuT, an oncogenic rat homolog of the receptor tyrosine kinase ErbB2, was transfected into the breast carcinoma cell line MCF7 using a vector based on the Tet-ON system. Upon incubation of the MCF7/NeuT cells with the antibiotic doxycycyline, upregulation of ErbB2/NeuT was achieved.

We investigated the signalling pathways that are involved in driving cells into a senescent state including their downstream effects on cyclins in cell cycle regulation. The results presented in this thesis show that upon ErbB2 upregulation significant changes in the cyclin dependent kinase inhibitor p21 occurred along with changes in the tumour suppressor genes p53 and PTEN and a cell cycle regulator, cyclin B2. Additionally, the knockdown of p21 in the presence of overexpressing ErbB2 reconfirmed that the MCF7 cells were able to maintain a proliferating state and escape senescence. This was determined by the upregulation of cyclin B2.

Moreover, a newly discovered oncogene was investigated for a potential role in senescence since it is suggested by literature to be involved in cell cycle arrest pathways. This oncogene, pituitary tumour transforming gene 1 (PTTG1) was found to be downregulated in overexpressing ErbB2 samples. When PTTG1 expression was tested in p21 knockdown samples it revealed an upregulation in expression, indicating that PTTG1 may have a role in senescence and p21 regulation.

Finally, the novel technique Raman micro-Spectroscopy was used to gain further insight into oncogene-induced senescence. Raman micro-Spectroscopy revealed changes in cytochrome *c* expression upon ErbB2 overexpression which was then reconfirmed by immunofluorescence and Western blotting where a higher level of cytochrome *c* expression was also detected in ErbB2 overexpressing cells.

In conclusion, the data presented in this thesis identified the up- or downregulation in expression of known senescent and cell cycle biomarkers which can be used to characterise the MCF7/NeuT cells in a senescent cell model. It also reconfirmed the involvement of p21 in senescence through gene knockdown studies by analysing cell cycle biomarker expression changes and also the effects of p21 on a newly discovered oncogene PTTG1. Finally, the novel technique, Raman micro-Spectroscopy, proved to detect changes between non-senescent and senescent cells indicating its use for further investigation into cancer diagnostics and research.