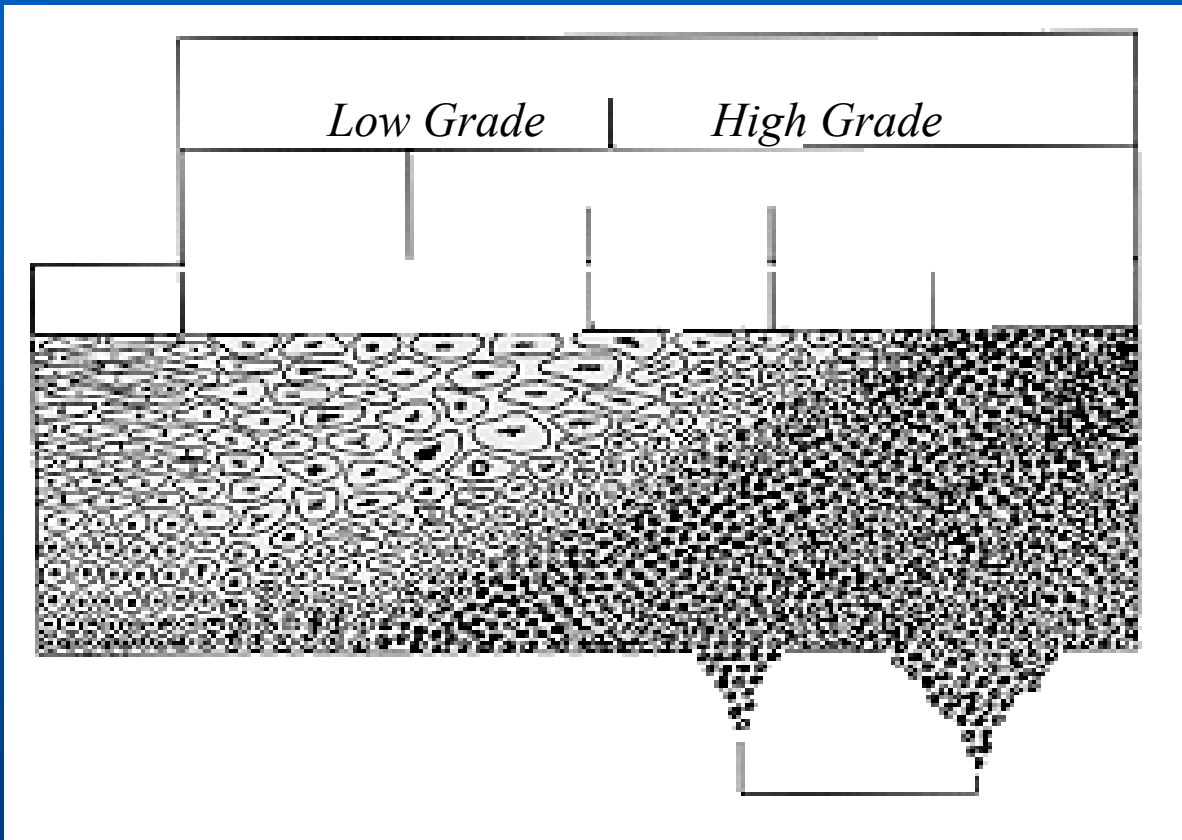
Project summaries

PDT & cervical dysplasia
PDT & adenovirus vector
PDT & angiogenesis

Photodetection of cervical intraepithelial neoplasia

- Screening, colposcopy, biopsy, treatment
- Optical biopsy
- See and treat

Cervical squamous carcinoma precursors



Performance of colposcopy for diagnosis of squamous intraepithelial lesions

First author	Se	Sp
Benedet,1976	0.99	0.53
Benedet, 1991	0.95	0.44
Crisforoni, 1995	0.97	0.35
Edebiri, 1990	0.87	0.67
Ferris, 1993	0.97	0.24
Javaheri, 1980	1.00	0.87
Lozowski, 1982	0.96	0.29
Seshadri, 1990	0.87	0.34
Stafl, 1973	0.99	0.26
Unweighted mean	0.95	0.44
Weighted mean	0.96	0.48

Mitchell MF et al ***Obstet Gynecol 93 : 462-70, 1999***

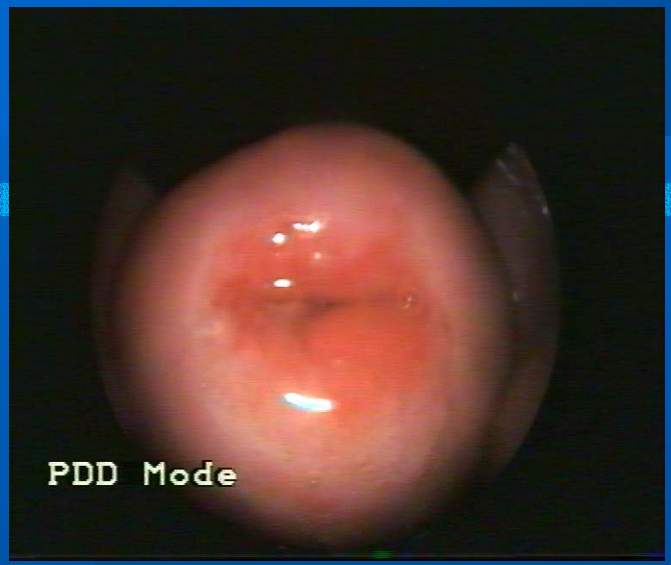
Photodetection of cervical intraepithelial neoplasia using 5-aminolevulinic acid-induced porphyrin fluorescence

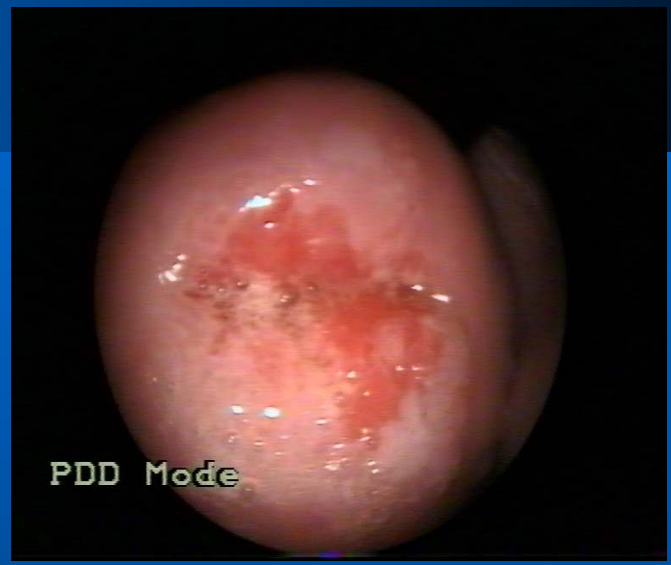
METHODS **Sixty-eight women attending our colposcopy clinic underwent a gynecologic examination, including cytology, human papillomavirus (HPV) testing, and colposcopy. They received 10 mL 0.5% or 1.0% 5-aminolevulinic acid (5-ALA) topically. After 30-360 minutes, real-time image analysis was performed, and spectra were obtained from 685 sites.**
RESULTS **Using 1% 5-ALA, fluorescence imaging after 60-90 minutes achieved similar sensitivity and specificity compared with colposcopy in detecting CIN with 94% and 51% versus 95% and 50%, respectively. However, the specificity was markedly improved by fluorescence spectroscopy, achieving 75%.**

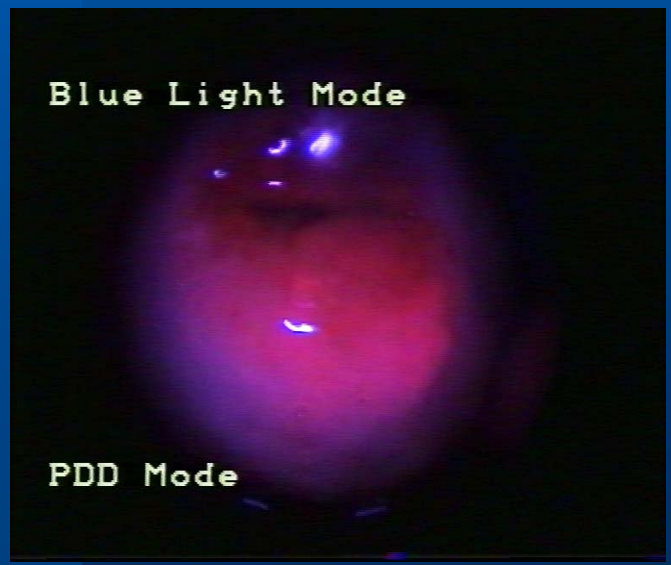
More info

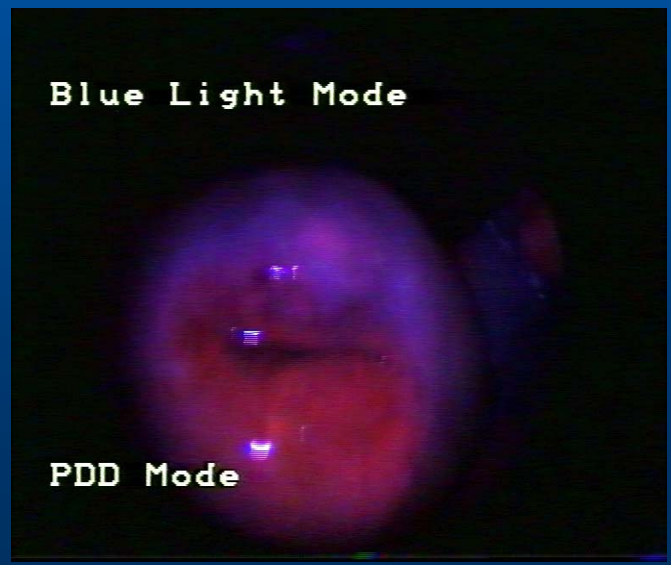
Peter Hillemanns et al Cancer 2000, 88 : 2275-82

Pat. 12, 1 % h-ALA in 0.9% NaCl, 2h 15 min application time

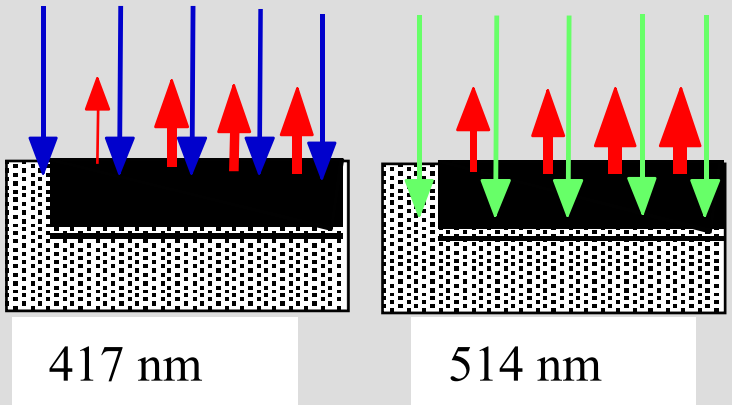








Principle of fluorescence imaging tumor depth profiling



Tumoral tissue with CIN
Normal tissue

Principle of fluorescence imaging tumor depth profiling

Homogenous exitation of the fluorochrome concentrated in the tumoral tissue at two different wavelenghts, corresponding to the absorption maxima of the fluochrome (417, 514 nm)

Detection at the emission maxima (610-720 nm)

Current status

- **Ongoing clinical study on PDT for cervical dysplasia**

General

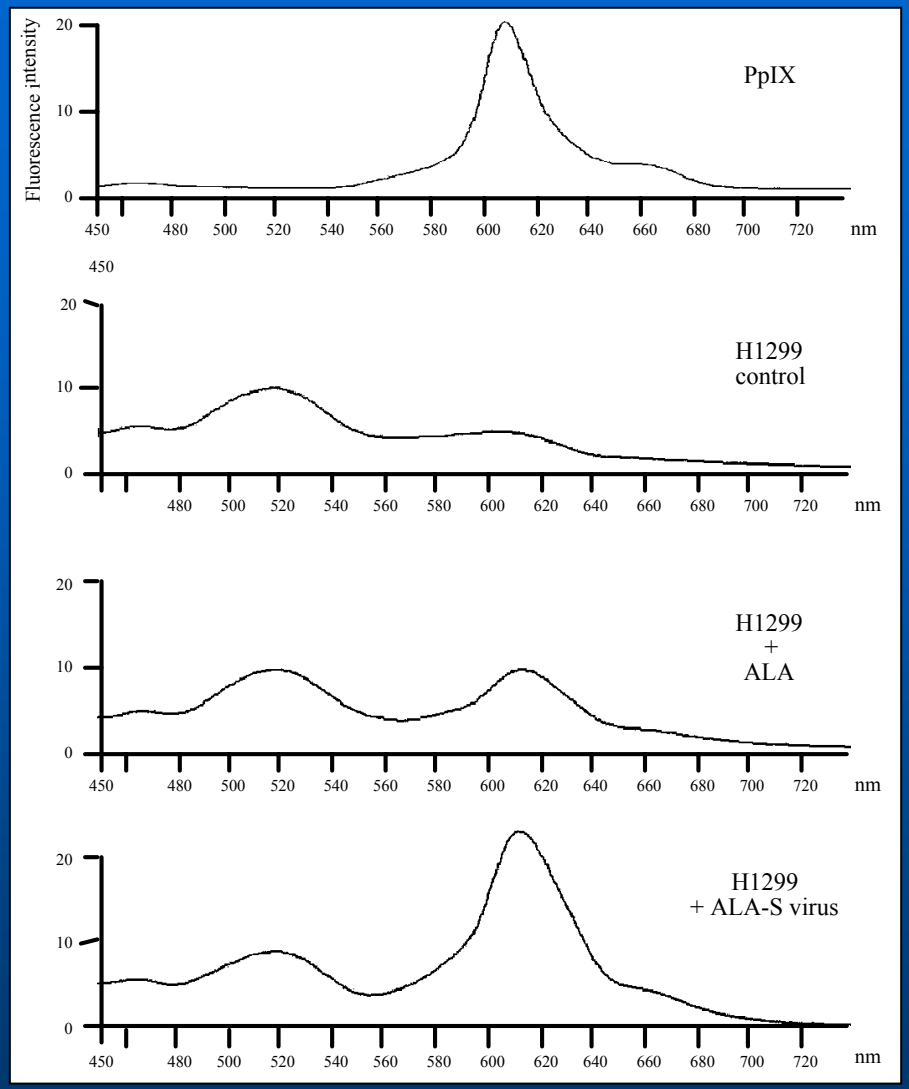
- Current ALA therapy lacks tumour specificity, impeding abdominal cavity photodynamic therapy
- Expression of the ALA-synthase mutant from a tumour specific promoter in a gene therapy vector should permit targeting of ALA production to tumour cells

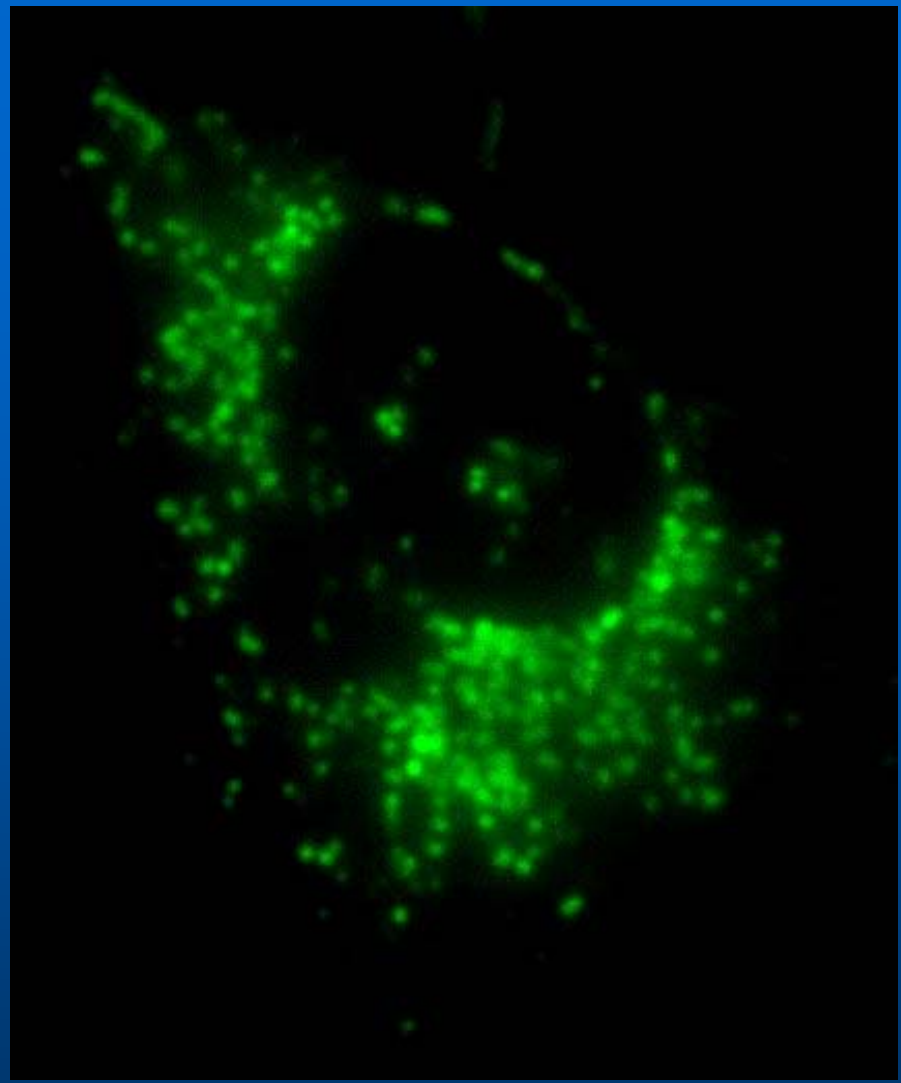
Specific aims

- To produce a broad host range adenovirus expressing ALA synthase from a tumour specific promoter
- Show that this virus efficiently enters cells
- Show that ALA synthase is expressed in a tumour specific manner
- Show that the virus selectively kills tumour cells in vivo following therapeutic irradiation.

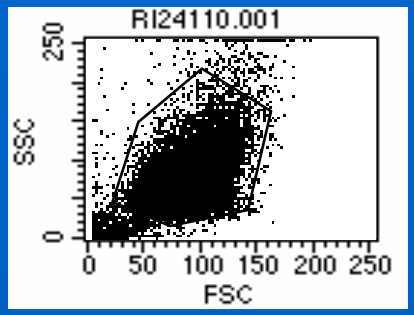
More info

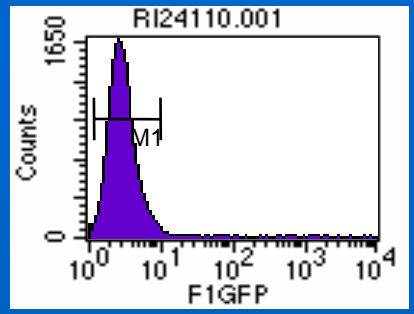
Gagnebin, J., Brunori, M., Otter, M., Juillerat-Jeaneret, L., Monnier, P., and Iggo, R. (1999). A photosensitising adenovirus for photodynamic therapy. Gene Ther *6*, 1742-1750

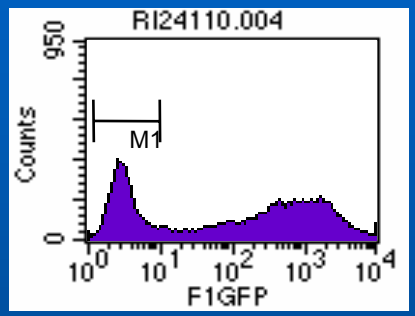

Fluorescence intensity
20
10
0
PpIX
450 480 500 520 540 560 580 600 620 640 660 680 700 720 nm
450
20
10
0
H1299
control
480 500 520 540 560 580 600 620 640 660 680 700 720 nm
20
10
0
H1299
+
ALA
450 480 500 520 540 560 580 600 620 640 660 680 700 720 nm
20
10
0
H1299
+ ALA-S virus
450 480 500 520 540 560 580 600 620 640 660 680 700 720 nm



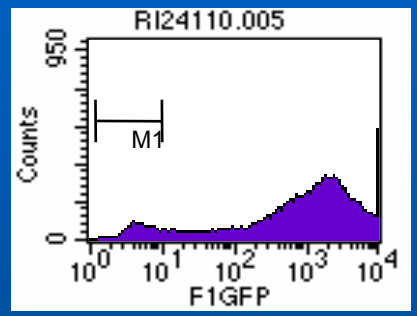
Cell line : HI299
Ad-CMV-GFP



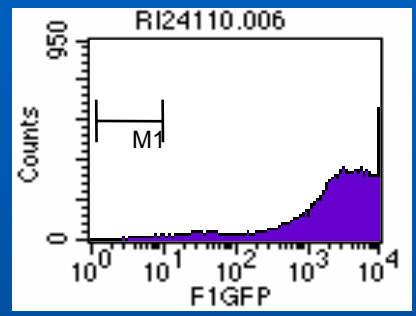




MOI 1

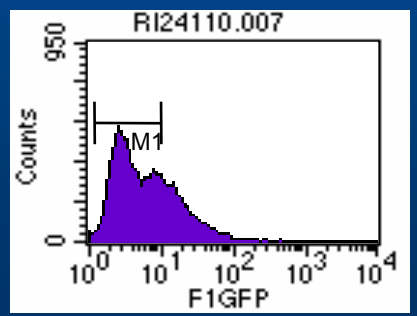


MOI 3

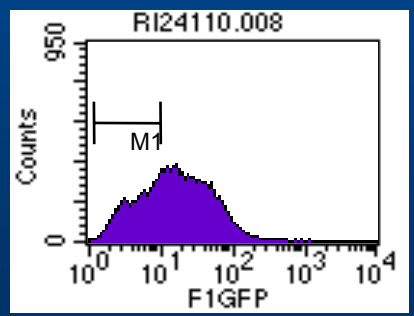


MOI 9

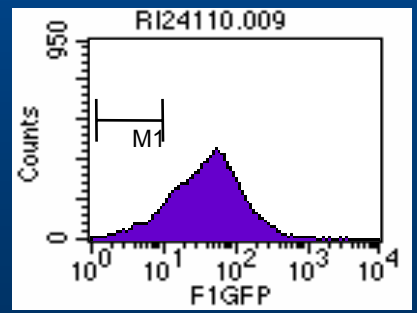
Cell line : HI299
Ad-E2F-GFP



MOI 1



MOI 3



MOI 9

Current status

- NuTu cells show low transduction rate (100 PFU/cell 60%transduction)
- Nude mice vs stable ALA-S cell line

General, PDT & Angiogenesis

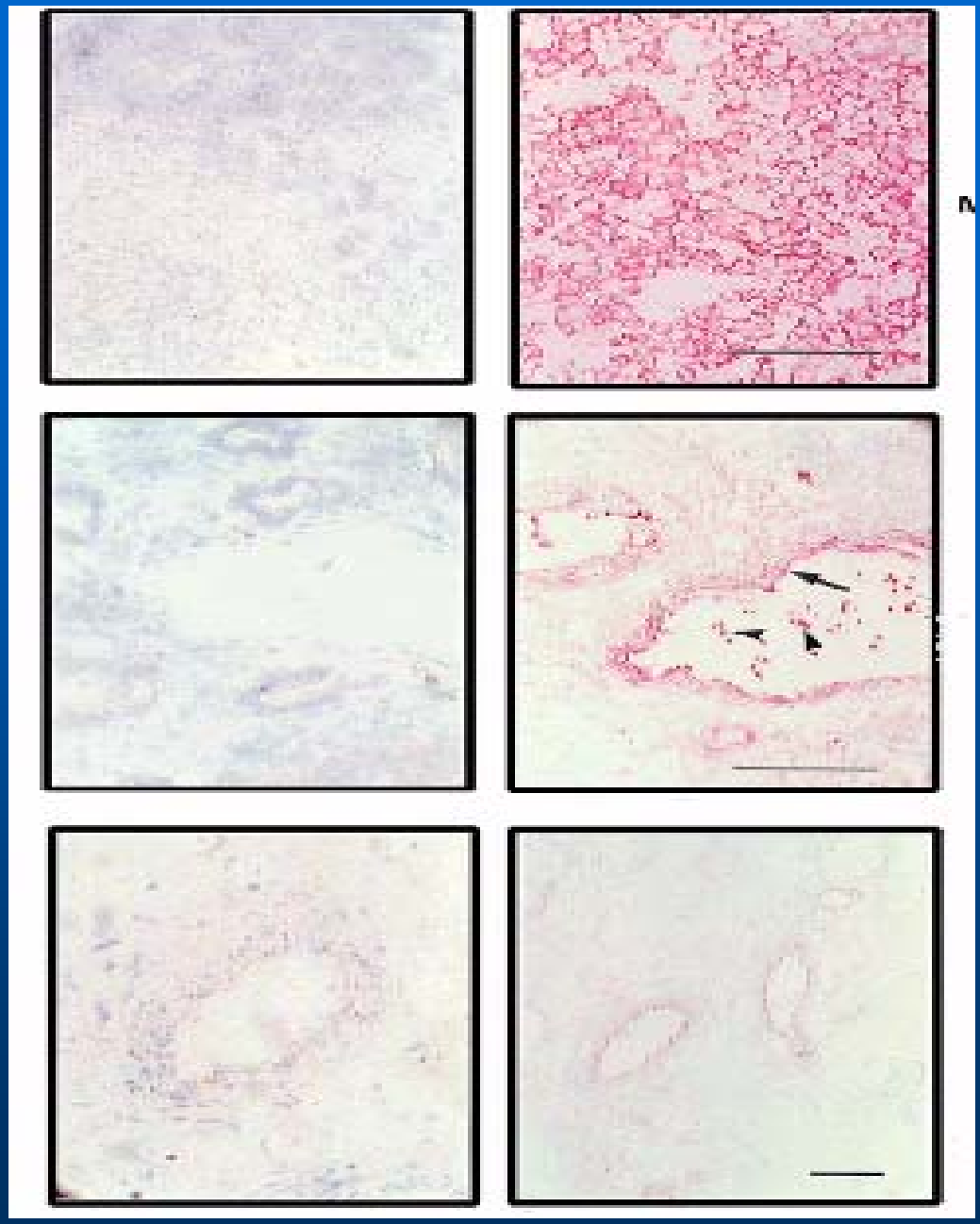
- By targeting specific angiogenesis markers and delivering fluorescent molecules to the vascular spread of ovarian cancer metastases early visualisation and treatment could establish a meaningful intervention to a much earlier point in the path of progression

Sky-7

- Disintegrins are a family of snake venom peptides of 60 to 70 amino acids in length and characteristically contain an RGD-motif that binds with high affinity to integrins. The disintegrin kistrin is unique since it shows binding specificity for $\alpha v \beta 3$
- Chimaeric integrin ligand termed SKI-7 which consists of the disintegrin kistrin fused to CD31. This setting allows coupling of drugs to the CD31 and it facilitates immunohistochemical detection of the chimeric molecule when it is bound to the target tissue

Specific aims

- To sythesize, characterize and purify the SKI-7 molecule
- To conjugate the targeting molecule to fluophores
- To test the binding and fluorescence characteristics of the designed molecule in vitro
- To perform photodiagnosis of ovarian cancer micrometastases in the rat animal model
- To run first preclinical tests of the fluorescence conjugates in ovarian cancer patients, establishing characteristic in vivo image information



Current status

- Production of (large) amounts of Sky-7
- Coupling to the photosensitizer