Supplementary Data

Recent advancements in imaging technologies have greatly enhanced the understanding of the influence of hormones on tumour progression in non-invasive conditions. Every imaging technology has its own pros and cons which can be circumvented by combining different imaging modalities, which increases the possibility of morphologically and molecularly characterizing the neoplastic disease. The main features of the most widely used in vivo imaging modalities are summarized here.

Radionuclides Imaging
Hormones, drugs or metabolite analogues (intermediaries of a biological process enhanced in the neoplasia) are labeled and generate two orthogonal positrons (e.g. $^{18}$F, $^{11}$C, $^{15}$O, $^{64}$Cu, $^{124}$I) or a single photon (e.g. $^{99m}$Tc, $^{111}$In, $^{123}$I, $^{125}$I, $^{201}$Tl, $^{77}$Br), the emissions are finally captured by a PET or SPECT scanner. Radioactive probes accumulate in body sites where a receptor is expressed or a biological process (e.g. proliferation, angiogenesis, apoptosis, inflammation) is occurring. With the help of imaging software the scanner localizes the radioactive source within the body in a highly-sensitive 2D or 3D reconstruction. Limited space resolution, relatively long acquisition time, difficulties in radiotracer development and production (a cyclotron is often needed), and the high costs of the instrumentation are the main limitations of this imaging modality.

Magnetic Resonance Imaging
This imaging modality is based on the resonance properties of nuclei oriented in a magnetic field temporarily exposed to a radiofrequency pulse direct in a perpendicular plane with respect to the magnetic field itself; the signal emission during the relaxation occurring after turning off the radiofrequency pulse gives information about the environment of the resonating nuclei. The outstanding feature of MRI is the very high resolution (25–100μm) which provides good contrast between several layers of soft tissues (e.g. complex tissue such as brain). In addition to morphological information, this method is also capable of providing functional parameters. The main limitation resides in the low sensitivity, long acquisition time and costly instrumentation.

Optical Imaging
Optical imaging can be applied to measure bioluminescent or fluorescent photon emissions even when the light source is coming from the inner tissues of a small animal (e.g. a mouse). Localization and measurement of the source requires an appropriate CCD-camera device. The molecular imaging of biological events can be achieved with fluorescent or bioluminescent probes, including reporter genes or molecules labeled with fluorescent moieties. This technology is fast, cost effective, very sensitive (especially for bioluminescence) with good resolution; however, optical imaging usually provides two-dimensional information with still limited three-dimensional possibilities.

Computed Tomography
CT is based on the different X-ray absorption of the various body structures due to their molecular composition; CT reveals the inner organ structure by providing high resolution (30–400μm) morphological information and is often combined with nuclear and optical imaging to obtain a detailed spatial localization of the radionuclide or light source.

Ultrasound imaging
Ultrasound wave pulses are reflected as echoes by tissues of different acoustic properties and the reflected pulses are received by the probe. Based on the Doppler effect, this imaging modality can reveal dynamic process such as blood flow in one organ. Contrast agents, such as microbubbles may enhance the echoes through resonance in the ultrasound beam. In the beginning ultrasound provided mainly morphological information with a good resolution (100–1000μm), nowadays ‘functionalization’ of ultrasound contrast agents allowed to selectively target the signal to a disease antigen or to a biological process such as ongoing inflammation or angiogenesis. Its cost effectiveness and short acquisition time make this emerging imaging technique more and more popular. The technique has limited (functional) quantification capabilities and is still heavily operator-dependent.