Diagnosis of malignant melanoma based on alterations in blood flow dynamics

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Biological oscillators

All biological systems are dynamical and *open* systems.

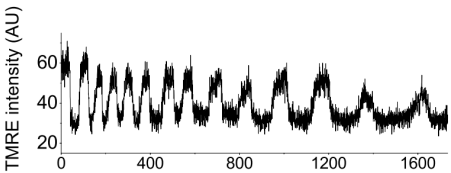
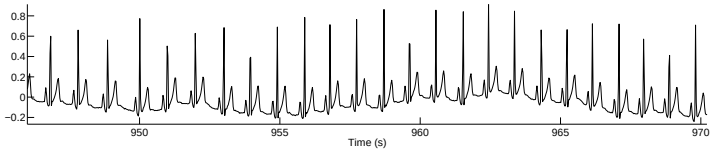


Figure: Kurz et al., PNAS, 2010.

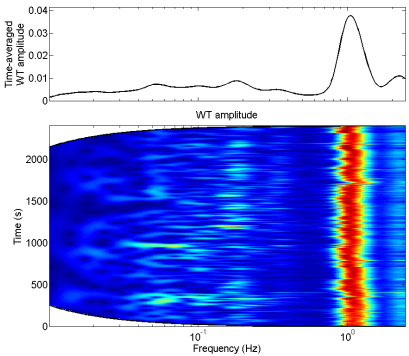
- Evidence of both deterministic and stochastic dynamics.
- Biological oscillations found on all scales.
- These have *functional relevance*.
- Shown to offer physiological advantages.
- Knowing the dynamics of a healthy biological system allows us to observe transitions to pathological state.
- Investigating the *interactions* between oscillators provides information about the system, e.g. causal relationships.

How do we model the dynamics of biological systems?

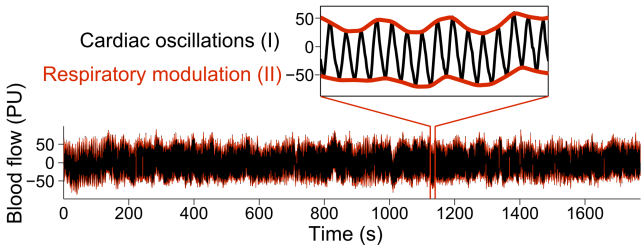
- We first need to *observe* the system over time → time series.



- Apply methods for time-varying data.
- Inspect for oscillations via transformation to the time-frequency domain.
- Then *extract* the dynamics of any observed oscillatory modes.
- Develop models which capture the main features, and allow *time-dependence*.



Example - Blood flow dynamics



- **Cardiac activity.** The pulse wave can be detected at all points in the body with a blood supply, most notably the wrist and neck.
- **Breathing**. Oscillations corresponding to the functioning of the lungs.

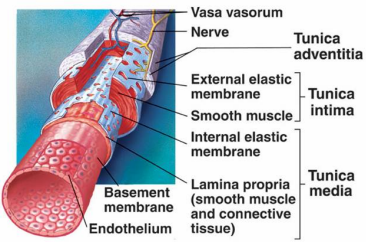
These are not the only oscillations that can be detected in blood flow.

Slower oscillations from other physiological processes have been detected at lower frequencies.

Blood flow dynamics – local effects

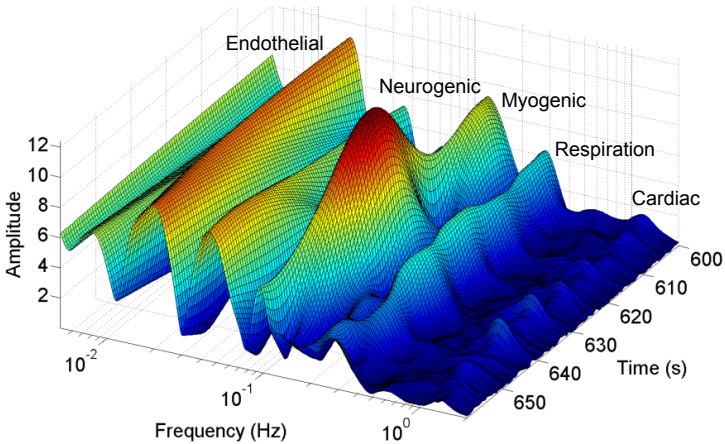
- **Myogenic activity**. Most vessels are surrounded by smooth muscle cells which can contract and relax to control the diameter of blood vessels depending on requirements.
- **Neurogenic activity**. Most vessels are in contact with nerves, which can stimulate a response in the smooth muscle cells, and alter the blood flow.
- **Endothelial activity**. Endothelial cells line all blood vessels and directly influence blood flow by releasing vasoactive substances such as nitric oxide (NO).

A combination of these effects manifests as oscillations in blood flow, as a consequence of *vasomotion*.



A. Stefanovska, IEEE Eng. Med. Biol. Mag., 26:25–29, 2007.

Blood flow dynamics



Characterizing these oscillations in healthy states has allowed their comparison in *pathological* states.

Main question:

What new information can we learn about cancer using biological oscillations?

- Cancer is a disease which affects millions of people worldwide.
- Invades and impairs many functions within the body.
- Prognosis generally worsens over time.
- Early diagnosis is crucial.

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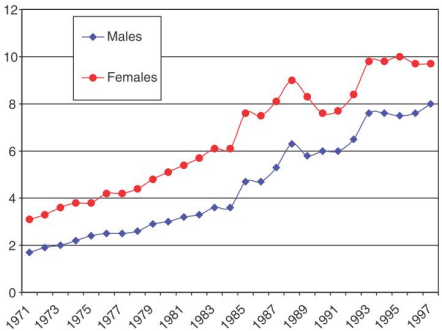
Research questions

1. Is blood flow dynamics altered in cancer?
   - Skin melanoma is an example where this question can be addressed by noninvasive measurements.
   - Compare between melanoma, atypical moles, benign moles, psoriasis and normal skin.
2. Are any observed differences large enough to be used in the development of a diagnostic test?
   - Differences provide functional information.
   - May also have clinical applicability.
3. What could be the possible reasons for these differences?

Melanoma

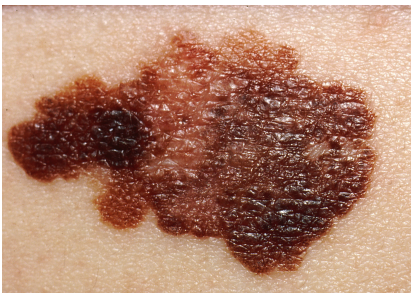


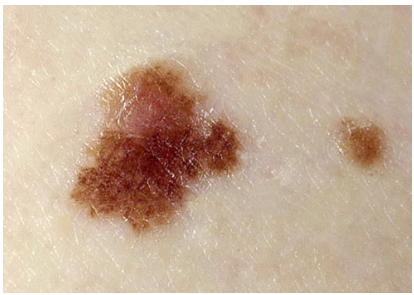
Instances per 100,000 of cutaneous malignant melanoma in the UK.

M. B. Lens & M. Dawes Br. J. Dermatol. 2003

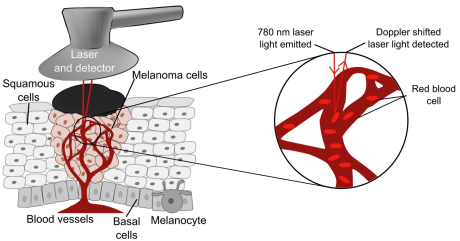
- Melanoma is the most dangerous skin cancer, causing 75% of all skin cancer related deaths.
- Requires early diagnosis and treatment for a positive long term prognosis.
- The gold standard of diagnosis is still invasive biopsy.

Noninvasive diagnosis?

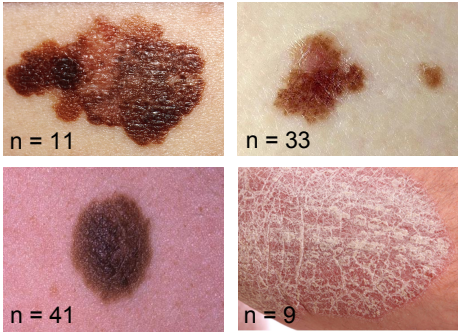




Blood flow dynamics in malignant melanoma



- Blood flow measured using LDF.
- 30 minutes.
- Compared different lesion types.
- 94 subjects.



Blood flow dynamics in malignant melanoma

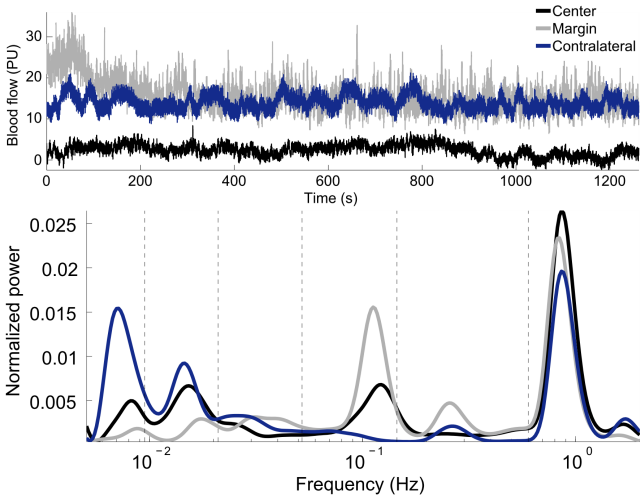
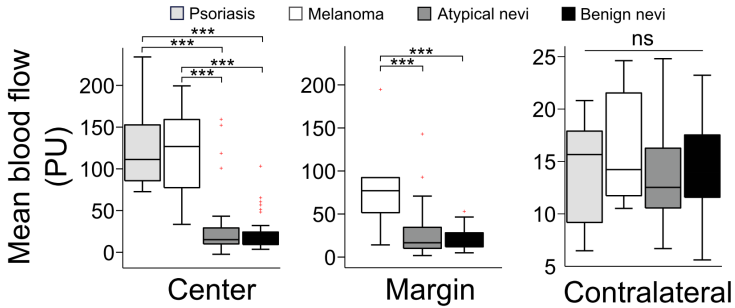
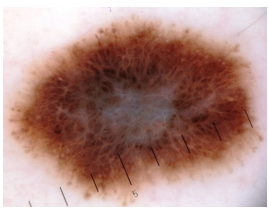


Figure: Example blood flow signals recorded in an atypical naevus using LDF, and their normalized wavelet power spectrum.

Average blood flow in malignant melanoma

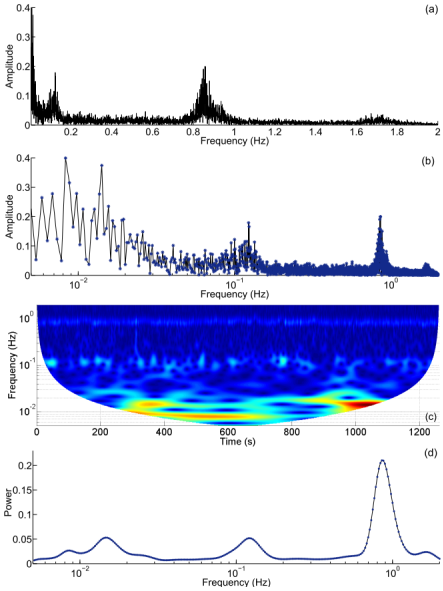


- Mean blood flow significantly higher in melanoma and psoriasis.
- Did not differ in contralateral skin.
- Using previous cutoff, specificity was 61%.
- No better than dermoscopy, another approach required.



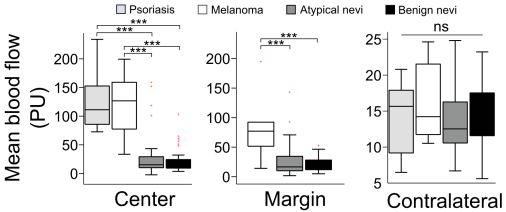
Analysing blood flow signals

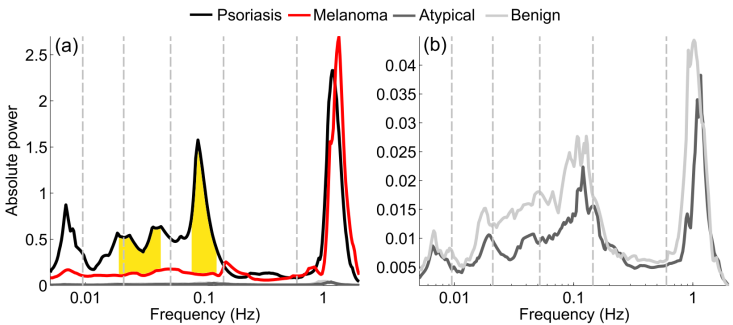
- We are investigating the dynamics of blood flow.
- Therefore, we need to investigate the frequency content.
- The Fourier transform does not allow for time-varying frequencies.
- The wavelet transform shows time and frequency information simultaneously.
- The wavelet transform can be averaged in time.
- This gives something analogous to the Fourier transform.
- However, the frequency resolution will be better at lower frequencies due to the logarithmic scale.



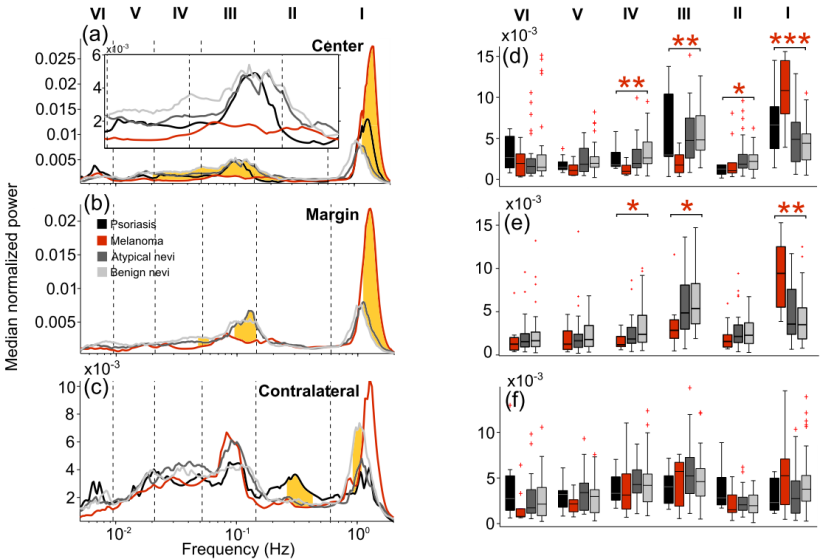
Blood flow dynamics in melanoma and psoriasis

- Mean blood flow does not differ between melanoma and psoriasis.
- But their dynamics does.
- Important to also consider frequency information.

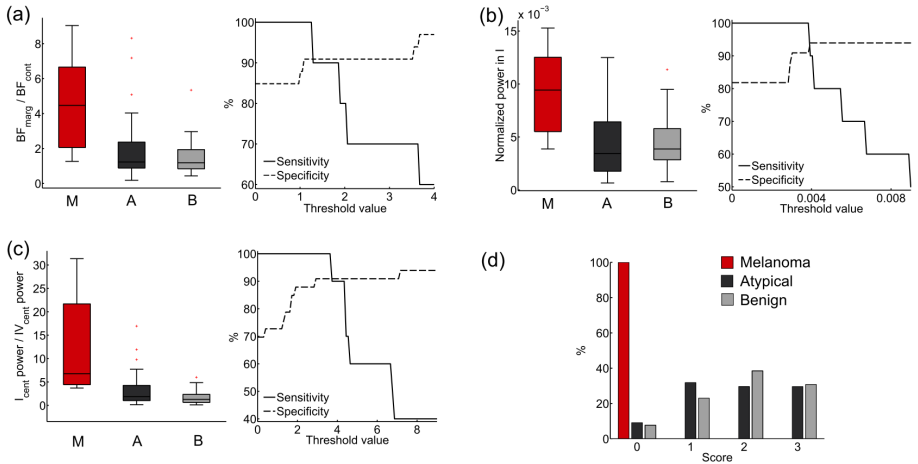




Blood flow dynamics in malignant melanoma



Non-invasive diagnosis in malignant melanoma



Diagnostic test results

Based on three parameters, a diagnostic test was developed with a sensitivity of 100% and a specificity of 90.9%.

Explaining the results

- Obtained a useful diagnostic test, but what can we learn from the results?
- Results suggest alterations in vessel function.

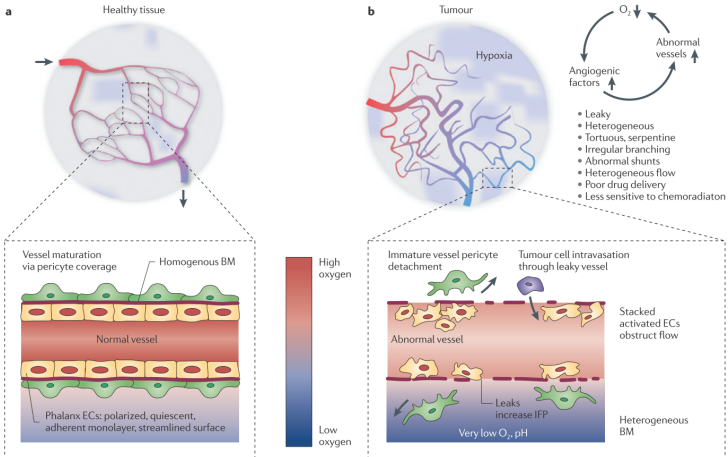


Figure: From Carmeliet, P. & Jain, R. K. *Nat. Rev. Drug Discov.* 2011.

- What else could contribute to the observed differences?

Biological oscillations in cancer

- There are many properties which vary between cancer types.
- Here, the focus is on universal properties.
- The main focus is two widely observed properties in cancer:

1. Tumours alter the surrounding vasculature via angiogenesis.
   - Hypoxia induces angiogenesis.
   - Vessels are different to normal vessels.
   - Normal regulatory processes may be impaired.
   - Thus, **blood flow dynamics** may be altered.
2. The metabolism of cancer cells is altered, e.g. the Warburg effect.
   - Glycolysis is upregulated even in the presence of oxygen.
   - Utilised in PET scanning.
   - Altered metabolism may directly affect the vasculature.

How alterations in metabolic oscillations could affect vessel function in cancer

- The metabolic switch to glycolysis results in a hostile microenvironment.
- This can reduce the reactivity of the vessels and induce angiogenesis.
- Normalization of the metabolism may improve the tumour microenvironment.
- Oscillations have been demonstrated in both glycolysis and the mitochondria.
- May be key to understanding the Warburg effect.

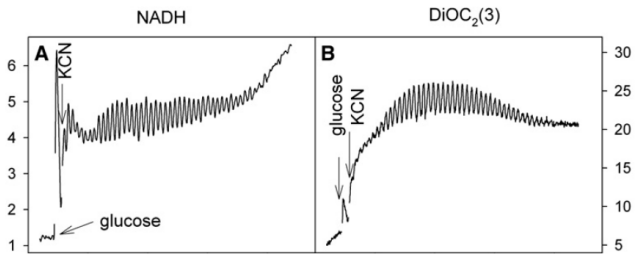


Figure: Oscillations in NADH and mitochondrial membrane potential observed with fluorescent dye in yeast cells. In this case, glycolytic oscillations drive those in the mitochondrial membrane. From Olsen et al. *Biophys. J.* 2009.

Conclusions

Answers to research questions:

1. Are blood flow dynamics altered in cancer?
   - Yes. Significant changes were observed in the normalized power in the cardiac, myogenic and neurogenic frequency intervals when comparing melanoma to atypical naevi, benign naevi, psoriasis and normal skin.

2. Are any observed differences large enough to be used in the development of a diagnostic test?
   - Yes. The effect size of the observed differences was such that a diagnostic test was developed which can distinguish noninvasively between melanoma and atypical naevi, the most difficult lesion to distinguish from melanoma.

3. What could be the reasons for these differences?
   - These differences likely arise from a combination of altered vascular structure via angiogenesis, and the effects of a hostile tumour microenvironment on the vasculature, including acidosis and hypoxia.
   - Acidosis results from altered energy metabolism, coupled with inadequate removal of waste.

Future Work

- Expand the melanoma diagnostic test data set.
- Develop a 'melanometer'.

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