

Cytokine networks in endometrial carcinogenesis

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Background

Endometrial cancer (EC) is the commonest gynaecological malignancy in the developed world and falls into Types I and II.

	Type I	Type II
% Cases	75-80%	20-25%
Classification	Endometrioid	Serous, clear cell
Premalignant stage	Hyperplasia	Rare
ER (+) ve status	Common	Rare
Grade	Low-medium	High
Outcome	Reasonable	Poor

Despite its role in immunoregulation/tumour behaviour, little is known about the cytokine microenvironment in EC. This study aimed to develop novel modelling approaches to clarify its role in carcinogenesis.

Methods

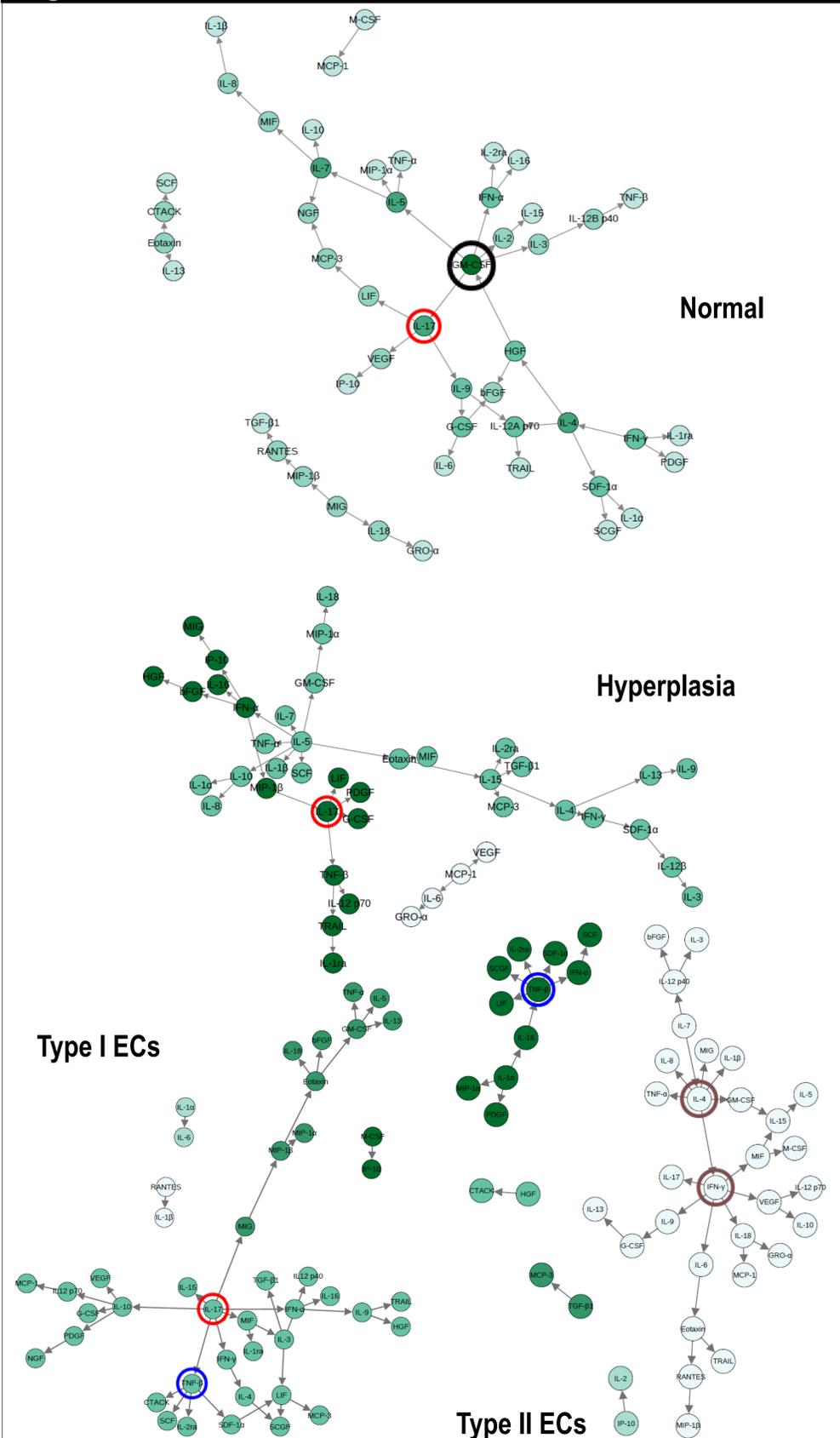
Frozen endometrial sample tissue lysates (38 normal, 25 hyperplastic, 97 cancers - 46 Type I/51 Type II) were profiled for 49 cytokines by multiplex immunoassay/ELISA. These included IL-1 α , IL-1 β , IL-1ra, IL-2, IL-2ra, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12 (p40)/(p70), IL-13, IL-15, IL-16, IL-17, IL-18, CTACK, eotaxin, bFGF, G-CSF, GM-CSF, GRO- α , HGF, IFN- α , IFN- γ , IP-10, LIF, MCP-1, MCP-3, M-CSF, MIP-1 α , MIP-1 β , MIF, MIG, bNGF, PDGF, RANTES, SCF, SCGF- β , SDF-1 α , TGF- β ₁, TNF- α , TNF- β , TRAIL and VEGF. All cytokine concentrations were standardised to total protein (Lowry assay).

Data were analysed by Kruskal-Wallis tests with Mann-Whitney-*U* tests *post hoc* applying FDR correction for multiple comparisons. Bayesian networks (which represent cytokines as nodes and their causal interactions as hierarchical directed edges) were constructed using an empirically validated sequence on *MetaCore*, *Predictionet*, *Matlab*, *R* and visualised in *Gephi*.

Results

Significantly different concentrations of 28 cytokines were noted across groups and absolute levels proved useful in differentiating between Type I/II cancers. These differences were reflected in cytokine interrelationships; distinct subnetworks with different nodal foci were seen across all groups (Figure 1). GM-CSF (black circle) appeared to play a major regulatory hub role in normal endometrium. Other hub node mediators were conserved: IL-17 (red circles) was consistently found in this capacity in all categories except Type II ECs, which instead favoured IL-4 and IFN- γ (maroon circles). Moreover, a central control role for agents such as TNF- β (blue circles) was only seen in cancers.

Figure 1



Hierarchical Bayesian networks for normal, hyperplastic and cancerous endometrium, highlighting prevailing hub nodes in the context of subnetwork structure.

Discussion

Endometrial carcinogenesis is associated with perturbations of intratumoural cytokine networks which, in the case of Type I malignancies, start to become apparent in premalignant hyperplasia. Interestingly these changes affected both the structural location and identity of hub (i.e. signal integrating) nodes within the network, and these differed across cancer subtypes. It is likely that such changes in inflammatory network architecture affect cancer cell proliferation and invasion as well as neoangiogenesis and escape from local immunosurveillance. Ongoing work is focussing on establishing concentration-dependent changes within the networks to highlight synergistic and antagonistic relationships with a view to identifying possible novel therapeutic targets as well as developing classifiers which aid in cancer prognostication based on tumour-specific cytokine signatures.

Acknowledgements

Funding: The authors are greatly indebted to Yorkshire Cancer Research and Wellbeing of Women for funding.