Surgery Open Science 18 (2024) 29–34

Contents lists available at ScienceDirect

Surgery Open Science

journal homepage: www.journals.elsevier.com/surgery-open-science

Research Paper

Differential cytokine and chemokine expression after ablation vs. resection in colorectal cancer liver metastasis

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ARTICLE INFO

Keywords:
Colorectal cancer
Liver metastasis
Radiofrequency ablation
Hepatectomy
Cytokine
Chemokine

ABSTRACT

Background: Surgical resection remains the main curative treatment for colorectal liver metastases (CRLM). Radiofrequency ablation (RFA) is increasingly employed for small, deep lying or otherwise inoperable lesions. However, RFA can induce pro-tumorigenic effects on residual tumor cells, hereby possibly promoting tumor recurrence. Contrastingly, post-RFA tumor debris as an antigen source can also generate anti-cancer immune responses. Utilizing this, current studies on combining RFA with immune therapy appear promising. Here, in an attempt to shed light on this controversy, cytokines involved in inflammation, (lymph)angiogenesis, immune cell recruitment and tumor cell invasion were investigated post-RFA versus post-resection in CRLM patients.

Methods: Cytokine and chemokine serum levels pre-operation, 4 h and 24 h post-operation were analyzed in CRLM patients undergoing RFA (n = 8) or partial hepatectomy (n = 9) using Multiplex immunoassays. Statistical analyses were performed between as well as within individual intervention groups.

Results: Post-RFA, significantly increased levels of acute phase proteins SAA1 and S100A8, IL-6, IL-1Ra, MIP3b (CCL19) and MMP9 were observed along with decreases in Fibronectin, MCP-1 (CCL2), and Tie-2. Post-resection, increased levels of PDGFbb, I309 (CCL1), Apelin, MIF, IL-1b and TNFα were seen. All p-values <0.05.

Conclusion: Pro-inflammatory responses mediated by different cytokines were seen after both RFA and resection, possibly influencing residual tumor cells and tumor recurrence. As both ablation and resection trigger inflammation and immune cell recruitment (albeit via distinct mechanisms), these data suggest that further research may explore combining immune therapy with not only RFA but also resection.

Key message: Analysis of patients' serum after radiofrequency ablation versus resection of colorectal liver metastases (CRLM) showed that these interventions trigger inflammation and immune cell recruitment, via different cyto- and chemokine pathways. This suggests a possible future strategy of combining immune therapy with not only ablative techniques but also with resection of CRLM.

Introduction

The liver is the most frequent site for distant metastases of colorectal cancer. Surgical resection remains the golden standard for potential curative treatment for colorectal liver metasastases (CRLM) [1,2]. If resection is possible, 5 year survival rates of 27–58 % are reached [1–5]. Unfortunately, at the time of diagnosis most hepatic lesions are not suitable for resection, considering factors such as extrahepatic involvement, future remnant liver function, and vascular structure involvement. In such cases, local ablative therapies such as radiofrequency ablation (RFA) may be deployed [6–8]. RFA is an image-guided modality using radiofrequency waves, which are converted into heat at the tip of an electrode using ionic agitation and friction. Tumor tissue adjacent to the electrode tip undergoes cell membrane destruction, protein denaturation and thermal coagulative necrosis [8–10].

While several studies have reported RFA to show comparable survival rates to surgical resection for small tumors, other studies judge it an inferior approach to hepatectomy [8,11–16] However, most studies have compared clinical outcomes of RFA for irresectable CRLM versus surgery for resectable CRLM. The question remains whether RFA could substitute resection for small resectable CRLM. Currently, a multi-center Phase-III randomized controlled trial is underway to compare surgical

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https://doi.org/10.1016/j.sopen.2024.01.005
Received 1 August 2023; Received in revised form 23 November 2023; Accepted 10 January 2024
Available online 14 January 2024

resection to thermal ablation in resectable CRLM smaller than 3 cm [17].

Considering the different modalities of RFA and resection, several research groups have investigated the change in local tumor area after these interventions. In RFA, the thermal damage and widespread cell destruction appears to trigger a local and systemic inflammatory response, with increased levels of IL-6 and acute phase proteins, as well as a local influx of immune cells [9,18–23]. A major limiting factor of RFA is whether it is able to achieve a sufficiently large ablation zone to realize complete tumor destruction. If not, then local and systemic cytokine and chemokine changes post-RFA could influence residual tumor cells. Indeed, RFA appears to promote a more aggressive phenotype in residual tumor cells. In fact, RFA is thought capable of promoting pro-tumorigenic processes such as proliferation, migration, EMT, increased stemness as well as neo-angiogenesis [9,21,24–28].

While a few studies have sought to compare the systemic cytokine response after RFA and resection and its impact on tumor recurrence, most focus has been on pro- and anti-inflammatory markers. In this study, we used a broad explorative approach in patients who underwent RFA or resection by including markers of pro- and anti-inflammation, (lymph)angiogenesis, immune cell recruitment, as well as factors involved in local tumor invasion. The aim was to investigate systemic cytokine changes in patients who underwent RFA or resection.

Methods & materials

Patient blood sample collection

Blood samples were collected from 17 patients with hepatic colorectal metastases who underwent either partial hepatectomy (9 patients) or radiofrequency ablation (8 patients) at the University Medical Center Utrecht in the Netherlands. To minimize the effect of resection size, only patients undergoing minor hepatectomy (< 4 liver segments) were included. Major hepatectomy patients were excluded. The samples were collected at 3 consecutive time points: directly pre-operatively, at 4 h and at 24 h post-operatively. The study protocol for collecting blood samples for research purposes was approved by the medical ethical committee of the University Medical Center Utrecht, the Netherlands and the Central Committee on Research Involving Human Subjects. Written informed consent was obtained from all patients. Blood samples were immediately transferred to the laboratory for further processing and storage at −80 °C.

Multiplex immunoassay

Serum levels of S100A8, SAA1, IL-6, TNFα, IL-1RA, IL-4, PDGFbb, MIP3b(CCL19), MCP1(CCL2), MIF, I309(CCL1), SDF1α(CXCL12), Tie2, VEGF, Apelin, MMP7, MMP9, TIMP1, Fibronectin, IL-1β, IL-8, IL-10, IL-18, IFNγ, TGFβ (LAP), PAI1, EGF, HGF, THBS1, GCSF and OPG were measured by Multiplex immunoassays at the Multiplex Core facility of the Center of Translational Immunology (UMC Utrecht, the Netherlands). Magnetic carboxylated polysterene microspheres (Luminex, Austin, TX, USA) were used to covalenty couple with the capture antibodies (50 μg/ml antibody per 6.25 × 10^6 microspheres) as previously reported [29–31] Positive (biotin coated) and negative controle microspheres (BD Biosciences) were included in each sample analysis. The assay procedures were performed as previously described [29–31]. To shortly summarize the assays, 50 μl of each sample was incubated with 10 μl microsphere suspension for 1 h. Afterwards, the suspensions underwent automated washing (sheath fluid, 0.5 % Tween20, 0.01% NaN3). Afterwards, each sample was incubated with 25 μl secondary antibody cocktail (8 μg/ml each) for 1 h. Samples again underwent automated washing. Then, samples were incubated with 25 μl of streptavidin R-phycoerythrin (BD biosciences, 25 ng per well) for 20 min. After a round of automated washing, samples were measured (in 100 μl HPE buffer). All described incubation steps were performed shielded from light, at room temperature, and under continuous shaking. Data acquisition was attained using a FlexMAP3D system (Bio-Rad) using xPonent 4.1 software (Luminex). For data analysis, Bioplex manager 6.1.1 (Bio-Rad) was used.

Statistical analysis

All statistical analyses were performed by SPSS 20 (IBM SPSS, Chicago, IL). For the statistical analyses, advice was obtained from the Julius Center for Data Science and Biostatistics (Utrecht, the Netherlands) for project support. Cytokines and chemokines are mediators that reflect changes in immune and inflammatory reactions. Collected data from such secreted factors have certain characteristics that are typical for cytokine expression data, which are challenging for statistical analysis. For instance, the data are often not normally distributed. A natural log transformation was applied to the data. Moreover, many factors are secreted at such low concentrations that they are too low to quantify. Therefore, the data often contained non-detectable values. For the non-detectable values, which were treated as "missing values", we used the Multiple Imputation method on the log transformed data. Missing values were replaced by imputed values. As it was known that these missing values were due to concentrations too low to quantify, the imputed values were set to fall between 0,0 and the known Lower Limit of Quantitation (LLOQ) of the analyses for each individual cytokine. After the Multiple Imputation method, the data was retransformed and reformatted as ratios of 4 h and 24 h compared to the pre-operative baseline. To further analyze the data, a linear mixed model was used. Within the model, pairwise comparisons were performed for the different consecutive time points within the intervention groups and between the intervention groups. A P value of <0.05 was considered statistically significant.

Results

Differentially expressed cytokine levels comparing post-Resection and post-RFA groups

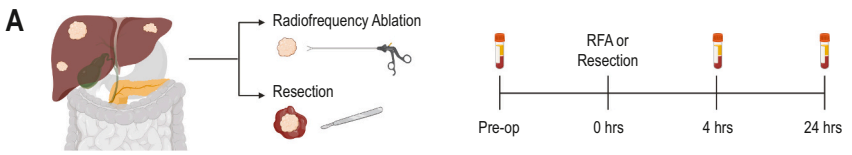
Post-resection, a marked increase in PDGFbb was seen compared to RFA (at 4 h and 24 h, respectively p-value = 0.023 and p-value = 0.006) (Fig. 1C). Apelin (APLN), an endogenous ligand for APJ receptors (APLNR), was increased at 4 h post-resection (p-value = 0.026). At 4 h post-resection, a considerable increase was seen in I309 (CCL1), involved in leucocyte (especially monocyte) recruitment [32] (p-value = 0.006) as well as for Macrophage migration inhibitory factor (MIF) (p-value = 0.037) (Fig. 1C).

In contrast to resection, post-RFA SAA1, an acute phase protein mainly produced by hepatocytes, was significantly elevated at 4 h (p-value = 0.014) (Fig. 1C). Also MIP3b (CCL19) was significantly increased at 24 h post-RFA (p-value = 0.030) (Fig. 1C). Both Fibronectin and Monocyte Chemoattractant Protein-1 (MCP-1, also known as CCL2) showed a marked decrease 24 h post-RFA (Fibronectin, p-value = 0.003; MCP-1, p-value = 0.005) (Fig. 1C).

Other plasma factors that were analyzed, namely acute phase protein S100A8, IL-1β, IL-4, IL-6, IL-8, IL10, IL-18, TNFα, IFNγ, TGFβ, SDF1α (CXCL12), Tie2, VEGF, MMP7, MMP9, TIMP1, PAI-1, EGF, HGF, THBS1, GCSF and OPG, were not significantly different when compared *between* the two intervention groups. Statistical analysis of cytokine levels of the different time points *within* the individual intervention groups showed further significant changes, mostly occurring within the post-RFA group.

Differential cytokine levels at consecutive time points within post-Resection group

At 24 h post-resection, elevated levels of pro-inflammatory IL-1b (4 h vs 24 h, p-value = 0.025) and pro-inflammatory tumor necrosis factor (TNF)-α (pre-op vs 24 h, p-value = 0.032) are observed (Fig. 1D, E).

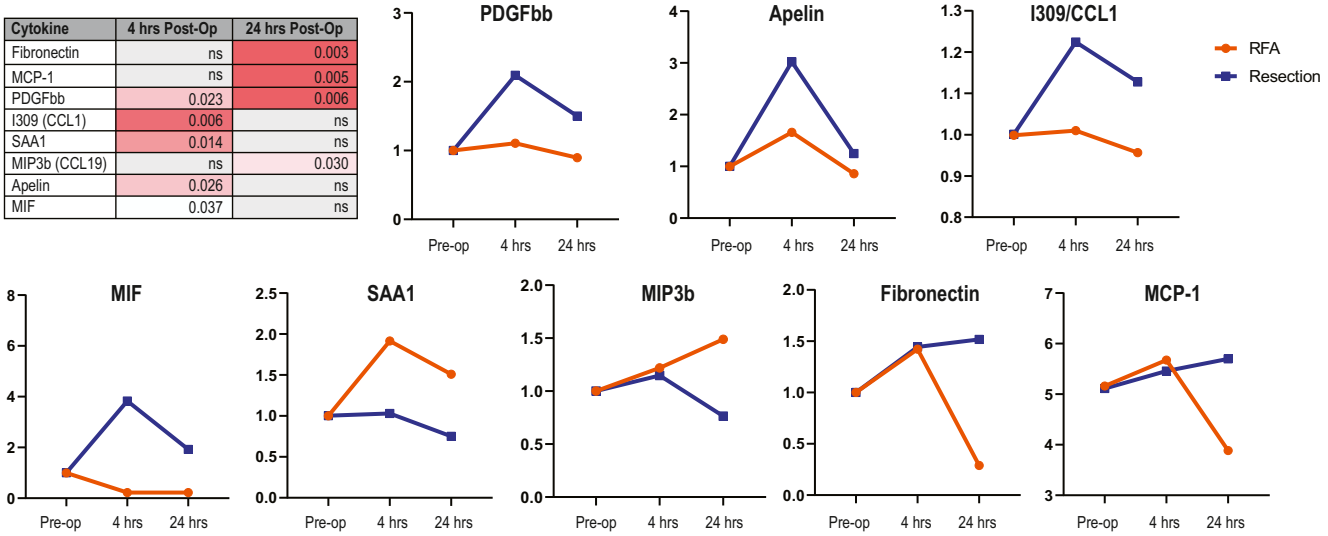


B

Pro-inflammatory S100A8, SAA1, IL-6, IL-1b, TNFα	**Anti-inflammatory** IL1Ra, IL-4, PDGFbb
(Lymph)angiogenesis Tie2, Apelin, VEGF, Ang2	**Tumor cell invasion** MMP9, TIMP-1, Fibronectin
Immune cell attractans MIP3b (CCL19), MCP-1, MIF, I309 (CCL1), SDF1α (CXCL12)	

C Differential expression comparing between RFA and Resection groups

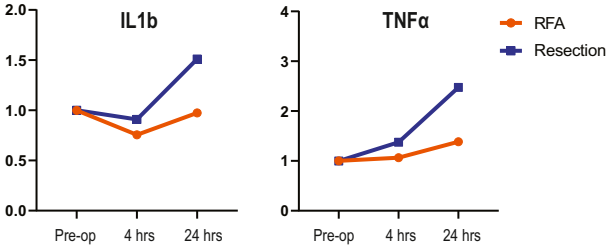
Cytokine	4 hrs Post-Op	24 hrs Post-Op
Fibronectin	ns	0.003
MCP-1	ns	0.005
PDGFbb	0.023	0.006
I309 (CCL1)	0.006	ns
SAA1	0.014	ns
MIP3b (CCL19)	ns	0.030
Apelin	0.026	ns
MIF	0.037	ns



D

Serum levels at time points within Resection group			
Cytokine	*Pre-Op vs 4 hrs*	*Pre-Op vs 24 hrs*	*4 hrs vs 24 hrs*
IL-1b	ns	ns	0.025
TNFα	ns	0.032	ns
Serum levels at time points within RFA group			
Cytokine	*Pre-Op vs 4 hrs*	*Pre-Op vs 24 hrs*	*4 hrs vs 24 hrs*
MMP9	0.001	ns	0.005
S100A8	ns	0.005	ns
Tie2	ns	ns	0.009
IL-6	ns	0.031	0.012
IL-1Ra	ns	0.054	0.022

E Serum levels compared within Resection group



F Serum levels compared within RFA group

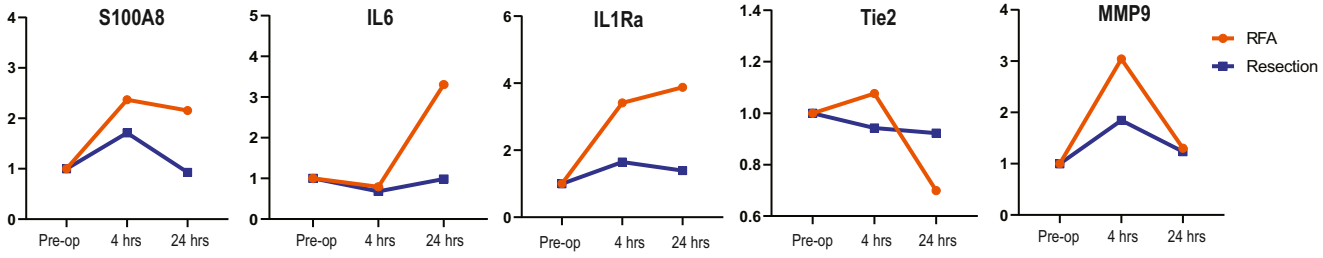


Fig. 1. A Schematic overview of the experimental set-up. At 3 consecutive time points (pre-operatively, 4 h and 24 h post-operatively), serum samples were taken of patients with colorectal liver metastases who underwent radiofrequency ablation (n = 8) or partial hepatectomy (n = 9). B Serum samples were used to analyze cytokine levels involved in pro-inflammatory, anti-inflammatory, chemotactic, (lymph)angiogenic and local tumor cell invasion processes. C PDGFbb, Apelin, I309 (CCL1), MIF, SAA1, MIP3b (CCL19), Fibronectin, and MCP-1 (CCL2) showed significant differential expression levels when comparing RFA vs Resection groups. Heat map of respective p-values comparing time points *between* intervention groups. Line graph visualization of serum level changes at consecutive time points in RFA (orange) and Resection (blue) groups. Plotted as ratio relative to pre-operative average. Ns = non significant. D Heat map of p-values showing significant differential cytokine expression when comparing time points *within* the intervention groups. Ns = non significant p-value. E When comparing time points within the Resection group, IL-1b and TNFα show significant increases at 24 h post-op. Plotted as ratio relative to pre-operative average. RFA (orange) and Resection (blue). F When comparing time points within the RFA group, S100A8, IL-6, IL-1Ra, and MMP9 show significant increased expression at 4 and 24 h post-op. Tie2 shows a significant decrease at 24 h post-op. Plotted as ratio relative to pre-operative average. RFA (orange) and Resection (blue).

Differential cytokine levels at consecutive time points within post-RFA group

Inflammation-related factors

At 4 h post-RFA, increased levels of acute phase protein S100A8 (pre-op vs 24 h post-op, p value = 0.005) was observed (Fig. 1D, F). At 24 h post-RFA, interleukin (IL)-6, an important mediator of the acute phase response that has both pro- and anti-inflammatory properties [33–35] was significantly increased (4 h vs 24 h, p value = 0.012). IL-1RA, the inhibitor of IL-1 and its pro-inflammatory signal [35], was markedly

increased (pre-op vs 24 h, p-value = 0.054, 4h vs 24 h, p-value = 0.022) (Fig. 1D, F).

Immune cell attractants

At 24 h post-RFA, a sharp decrease was seen for the Monocyte Chemoattractant Protein-1 (MCP-1, also known as CCL2) post-RFA (pre-op vs 24 h p-value = 0.014 and 4 h vs 24 h p-value = 0.001) (Fig. 1C).

(Lymph)angiogenic factors

At 24 h post-RFA, angiotensin receptor Tie2 (also known as TEK receptor tyrosine kinase) showed a pronounced decrease (4 h vs 24 h, p value = 0.009) (Fig. 1D, F).

Local tumor cell invasion factors

At 4 h post-RFA, matrix metallopeptidase 9 (MMP-9) shows a sharp increase (pre-op vs 4 h, p-value = 0.001) with a sharp decline at 24 h (4 h vs 24 h, p-value = 0.005) (Fig. 1D, F).

Discussion

RFA and Resection induce pro-inflammatory responses, with different cytokine mediators

In this study comparing systemic cytokine levels after RFA and resection, several cytokines were observed to be significantly differentially expressed post-intervention. When compared directly to the resection group, post-RFA showed an increase of acute phase protein SAA1 as well as a decrease in fibronectin. Fibronectin is involved in various processes such as wound healing, cell migration and is often decreased in acute inflammation [36,37]. When comparing cytokine levels at consecutive time points within the RFA group, significantly increased levels of acute phase protein S100A8 and pro-inflammatory IL-6 were observed. Indeed, increased levels of acute phase proteins and IL-6 have been consistently reported post-RFA [19–24,28]. Moreover, a sharp increase of anti-inflammatory IL-1Ra (receptor antagonist of pro-inflammatory cytokine IL-1) [38] was seen at post-RFA.

Post-resection, different pro-inflammatory mediators were observed. Comparing cytokine levels at different time points within this group, increased levels of pro-inflammatory IL-1b and TNFα were observed. Moreover, increased levels of PDGFbb were seen. While the role of PDGFbb in inflammation has not been completely elucidated, in patients experiencing sepsis PDGFbb levels negatively correlated with levels of pro-inflammatory cytokines IL-6, IL-1b, IL-8 and TNF-α [39]. Therefore, it appears both RFA and resection induce inflammatory responses, however with different mediators. RFA induces a response marked by increased levels of IL-6 and acute phase proteins SAA1 and S100A8 as well as decreased anti-inflammatory IL-1Ra, while post-resection shows increased levels of IL-1b, TNFα and PDGFbb.

Leucocyte recruiting chemokines increased post-resection, not post-RFA

Due to its nature, RFA generates considerable cell debris which provides a possible antigen source for immune system activation [18]. Directly post-RFA, several studies found a large influx of immune cells at the periphery of the ablated region [18,19,40–42]. Paradoxically, in the current study CCL1 and MIF, involved in monocyte and macrophage recruitment, were significantly increased post-resection compared to post-RFA. Moreover, potent leucocyte recruiting chemokine MCP-1 (CCL2) was decreased post-RFA. Therefore, we did not observe a cytokine pattern linking RFA to more immune cell recruitment compared to resection, but the opposite. However, the strong immune cell influx observed post-RFA in other studies could be mediated by chemokines not included in this study.

Possible pro-tumorigenic effects on residual tumor cells after RFA: IL-6 and MMP9

A technical disadvantage of RFA is the difficulty to achieve complete ablation of all tumor cells. As a result, residual tumor cells could contribute to the highly variable recurrence rates observed after RFA [8,9,11]. Therefore, recent studies have focused on whether RFA could stimulate tumorigenesis and tumor growth, not only in local residual tumor cells but also those situated in distant organs. Indeed, residual tumor cells after RFA appear to exhibit a more aggressive phenotype and RFA might even stimulate distant tumor growth [9,19,21,24,25]. RFA has been observed to promote proliferation, migration, EMT, increased stemness as well as angiogenesis [24,26–28,43]. In our study, increased level of IL-6 post-RFA could provide a strong pro-tumorigenic signal to residual tumor cells. Post-RFA increased expression of IL-6 has been shown to induce proliferation, migration and invasion in residual tumor cells after insufficient ablation as well as promote the polarization of pro-tumor macrophages [21,24,28,33,44].

In this study, a strong increase in metalloproteinase-9 (MMP-9) was observed post-RFA. MMP-9 plays a crucial role in cancer progression due to its involvement in ECM remodeling and neovascularization [45]. Studies have shown that incomplete RFA induces increased expression of MMP-9 as well as VEGF, the latter linked to (lymph)angiogenesis [24,46]. In this study, aside from a decrease in angiotensin receptor Tie2 post-RFA, no significant changes in VEGF, Angiotensin-2 or other (lymp) angiogenic factors were observed. However, an increase in CCL19 (MIP3b) was observed post-RFA, while a decrease was seen post-resection. The CCL19/CCL21/CCR7 axis is known to have possible pro-tumorigenic and pro-(lymph)angiogenic effects by inducing VEGF-A/C/D expression. Activation of CCR7 on cancer cells also induces EMT and tumor migration [47–49]. Therefore, it would be interesting in future studies to investigate VEGF-C/D, CCL21 and CCR7 levels post-RFA and resection.

Possible pro-tumorigenic effects on residual tumor cells after Resection: IL-1b, CCL1, MIF, Apelin

Interestingly, while recent studies have focused on pro-tumorigenic changes post-RFA, cytokine changes after resection could also influence residual tumor cells. Pro-inflammatory IL-1b which showed a significant increase post-resection, can also exert pro-tumorigenic effects on lung and liver metastases [38].

Furthermore, both CCL1 and MIF were increased post-resection. CCL1, expressed in various cancer types, can stimulate tumor cell proliferation and migration, as well as lymph node metastases [32]. MIF is a key inducer of inflammatory cytokines TNF-a, IL-1 and IL-6 and over-expressed in many solid tumors. In-vitro inhibition of MIF showed reduced tumor growth, progression and neo-angiogenesis [50,51].

Post-resection showed increased Apelin levels, a ligand for Angiotensin-like-receptor 1 (APJ). In various solid tumors, increased expression of Apelin and APJ has been observed. The Apelin/APJ axis appears to be involved in tumor cell proliferation, migration, metastasis, angiogenesis. Moreover, Apelin appears to inhibit tumor cell apoptosis and enhances resistance to especially anti-angiogenic and apoptosis-inducing drugs [52].

Platelet derived growth factor homodimer bb (PDGF-bb) was also increased post-resection. PDGF-bb is shown to promote lymphangiogenesis and lymphatic metastases in NSCLC [53], while showing anti-tumor properties in colorectal cancer [54].

RFA and Resection: combination with immune therapy?

Recently, studies have focused on whether the tumor debris-inducing and antigen-presenting nature of RFA tissue destruction could be combined to amplify targeted immune therapy [55]. In fact, the combination of RFA with various forms of immune therapy was shown to bolster the

anti-tumor effect of both treatments [56–58].

In this study, both RFA and resection showed increased pro-inflammatory responses post-intervention. Moreover, post-resection showed increased levels of immune cell recruiting factors compared to RFA. Therefore, in patients where no complete tumor resection was achieved or where several inoperable lesions remain in place, combining immune therapy with resection might provide an interesting approach in future studies to target residual tumor cells.

Limitations

There are several limitations to the current study. First, it was restricted to a small data set with 8 patients for the RFA group and 9 patients for the resection group, at three time points. However, as other studies also showed changes in various cytokine levels at 12-24 h post-intervention [20–23], we believe the chosen time points were a prudent approach.

Second, an inherent difficulty of cytokine data analysis is cytokine levels that are below the limit of detection. In this study, we implemented a multiple imputation approach for the statistical analysis of these undetectable values.

Thirdly, to more accurately compare RFA and resection, in this study only patients undergoing minor hepatectomy (< 4 liver segments) were included. As standard clinical practice, liver function was monitored post-operatively and none of the patients developed impaired liver function, as defined by the International Study Group of Liver Surgery (ISGLS) criteria for posthepatectomy liver failure. Therefore, we do not expect this to have significantly influenced our cytokine and chemokine arrays. However, it cannot be definitively excluded as a possible confounding factor.

Conclusion

In patients with colorectal liver metastases, after both RFA and resection pro-inflammatory systemic responses were observed, mediated by different secreted factors. For both interventions, these systemic cytokine responses could influence residual tumor cells and promote tumor recurrence. While recently the combination of RFA with immune therapy appears promising, the current study showed resection also triggers inflammation and immune cell recruitment (albeit via different cyto- and chemokines). These data suggest further exploration into whether combining not only RFA but also resection with immune therapy might offer future treatment strategies.

Abbreviations

CRLM	Colorectal cancer liver metastases
CCL1/I309	Chemokine (C-C motif) ligand 1
ECM	Extracellular matrix
EGF	Epidermal growth factor
GCSF	Granulocyte colony-stimulating factor
IFNγ	Interferon gamma
HGF	Hepatocyte growth factor
IL	Interleukin
IL-1β	Interleukin 1β
IL-1Ra	Interleukin-1 receptor antagonist
MCP-1	Monocyte chemoattractant protein-1 (also known as CCL2)
MIF	Macrophage migration inhibitory factor
MIP3β	Macrophage Inflammatory Protein 3 Beta (also known as CCL19)
MMP	Matrixmetalloproteinase
OPG	Osteoprotegerin
PAI1	Plasminogen activator inhibitor-1
PDGFbb	Platelet-derived growth factor (BB isoform/dimer)
RFA	Radiofrequency ablation
S100A8	S100 calcium-binding protein A8
SAA1	Serum amyloid A1
SDF1α	Stromal cell-derived factor-1 α (also known as CXCL12)
TGFβ (LAP)	Transforming growth factor beta (also known as latency-associated peptide)
THBS1	Thrombospondin 1
Tie2	Tyrosine-protein kinase receptor Tie-2 (also known as Angiopoietin-1 receptor)
TIMP1	Tissue inhibitor of metalloproteinases 1
TNFα	Tumor necrosis factor alpha
VEGF	Vascular endothelial growth factor

Ethical approval statement

The study protocol for collecting blood samples for research purposes was approved by the medical ethical committee of the University Medical Center Utrecht, the Netherlands and the Central Committee on Research Involving Human Subjects. Written informed consent was obtained from all patients.

Credit authorship contribution statement

Nicola Frenkel: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. **Susanna Poghosyan:** Data curation, Formal analysis, Investigation, Methodology, Writing – original draft, Writing – review & editing. **Jan Willem van Wijnbergen:** Data curation, Formal analysis, Investigation. **Inne Borel Rinkes:** Conceptualization, Funding acquisition, Investigation, Project administration, Resources, Supervision, Writing – original draft, Writing – review & editing. **Onno Kranenburg:** Conceptualization, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Writing – original draft, Writing – review & editing. **Jeroen Hagendoorn:** Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Writing – original draft, Writing – review & editing.

Declaration of competing interest

The author(s) declare no competing interests.

Acknowledgements

This work was supported by the KWF Dutch Cancer Society/Koningin Wilhelmina Fonds Kankerbestrijding (grant no. UU2014-6904 to J. H.) and the Alexandre Suerman Young Talent Stipend (N.F.). Fig. 1A and B were created with BioRender.com.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.sopen.2024.01.005.

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