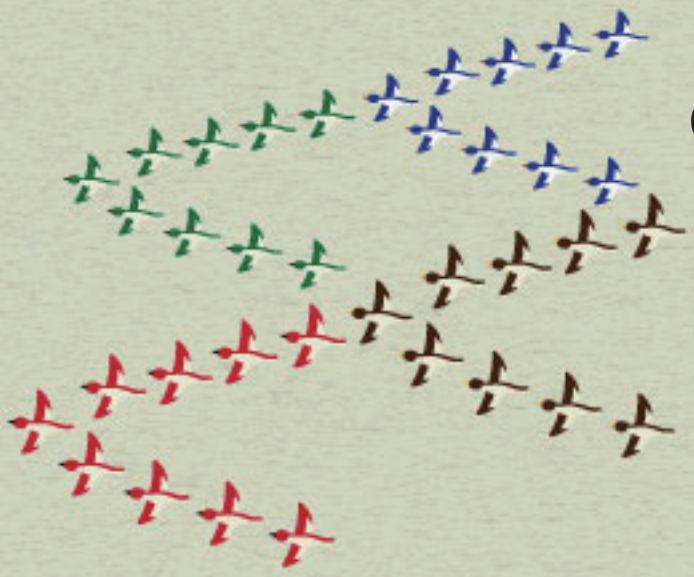

QST-M-14

第 3 期国際オープンラボラトリー資料集
（第 3 期 IOL）

Records of the 3rd Term
International Open Laboratory
(3rd IOL)

2018 年 9 月

September, 2018

放射線医学総合研究所

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目次

１．第３期 IOL 委員会報告書

第 3 期「放医研国際オープンラボラトリー」審査結果報告

放射線医学総合研究所理事長殿、
2015 年 5 月 1 日

今般、事務局の依頼を受けて、第 3 期「放医研国際オープンラボラトリー」（以下、本事業と呼ぶ）の課題審査を実施したので、その結果を報告する。この結論は、委員 5 人の総意である。

1. IOL 委員会構成

IOL 選考・評価委員会（以下、本委員会と呼ぶ）は以下のメンバーから構成される。

IOL 選考・評価委員会（順不同）

- 簇野嘉彦（東京工業大学名誉教授）
- 丹羽太貫（福島県立医科大学特命教授）
- 榮　武二（筑波大学教授）
- 河野俊之（東京工業大学教授）
- 柴田裕実（大阪大学産業研究所特任研究員、委員長）

事務局

放射線医学総合研究所企画部国際連携推進室
（村上健、藤田敬、伊藤悦子）

2. 募集、及び採択の経緯

本事業は以下のようなスケジュールで募集が行なわれた。（この部分、事務局記入）

- 2014 年 12 月 15 日：理事長決定で本事業を立ち上げ
- 同 12 月 25 日：公募開始
- 2015 年 2 月 20 日：公募締め切り

その結果、以下の 8 課題の応募があったことが、事務局より報告された。また、事務局で確認した結果、全ての応募課題が、募集要領に書かれた応募の条件は満たしていることが報告された。

コア番号	コア名	コアリーダーJ	コアリーダーF
15FL-A01	Development of strategies for enhancing efficiency of heavy ion radiotherapy	LEE, Younghyun, 28? ylee@nirs.go.jp	NICKOLOFF, Jac, 58?
15FL-A02	Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam	KONISHI, Teruaki, 39 tkonishi@nirs.go.jp	WANG, Jun, 38
15FL-A03	CASCET (Cancer Stem Cell Team)	VAREZ, Guillaume, 34 vares@nirs.go.jp	DURANTE, Marco, 50

15FL-A04	Proteomic analysis of protein modifications induced by C-ion irradiation	FUJITA, Mayumi, 39 mafujita@nirs.go.jp	WINK, David, 57
15FL-A05	Radiation damage mechanism at molecular level approached with physichemical technologies	KODAIRA, Satoshi, 35 koda@nirs.go.jp	BARILLON, Remi, 49
15FL-A06	Heavy ion radiotherapy in hypoxia	HIRAYAMA, Ryoichi, 37 hirayama@nirs.go.jp	SCIFONI, Emanuel, 45
15FL-A07	Impact of heavy ion irradiation on stem cells and tissue regeneration	SHIMOKAWA, Takashi, 44 takshi@nirs.go.jp	KASPER, Maria, 36?
15FL-A08	Whole gamma imaging	YAMAYA, Taiga, 41 taiga@nirs.go.jp	PARODI, Katia, 40

応募資料に基づき、以下のような日程で審査を行った。

- 2 月 26 日～3 月 1 日：応募書類のコピーを、事務局から各委員に送付。
- 3 月 2 日～3 月 19 日：委員の間でメールによる意見交換を実施。
- 3 月 20 日：委員 5 人及び事務局が東京八重洲ホール 102 会議室に集まり、1 回目の IOL 選考委員会を開催。
- 3 月 21 日～4 月 2 日：各委員が書類審査の結果を事務局に送付、事務局はそれらをまとめた書類審査結果の案を各委員に送付。
- 4 月 3 日～4 月 6 日：書類審査結果案を全員が承認。それに基づきヒアリングを実施する 4 課題を決定。
- 4 月 3 日～4 月 8 日：申請者、および委員のスケジュール調整を行い、ヒアリング日程を決定。
- 4 月 30 日：放医研、重粒子治療推進棟地下セミナー室 2 においてヒアリング実施。引き続き、2 回目の IOL 選考委員会を開催。採択案を決定。

3. 採択に関する方針

本委員会の委員は全員外部委員であるため、基本的に申請書及びヒアリングの内容に基づいて判断を行っている。申請書に書かれていない内容（記入されていないこれまでの成果など）や、それ以外の情報（所属部署がどこかなど）は基本的に考慮していない。若干の情報は、必要に応じて事務局より説明を受けたが、結論を左右するほどの影響はなかった。

本委員会としては、本事業の趣旨は、公募要領に書かれている通り、以下のような点にその真髄があると理解している。

✓ 海外交流の促進を通じて、放射線に関する様々な分野における「高度な研究シーズの創出」を推進すること

- ✓ 海外のアクティブな研究者との交流を通じ、次期放医研の中核を担いうる革新的な研究テーマの創出
- ✓ 失敗を恐れない自由闊達な研究の着想を積極的に支援

従って研究内容や計画性はもちろん重要であるが、それと同等に、あるいはそれ以上に、現在の研究内容から飛躍する姿勢、新しい分野にチャレンジする内容であることが重要であるというのが、全委員の一致した認識である。したがって、提案内容の新規性や他分野との交流について高い評価を与えた。

また、「将来のシーズ探し」ということは、当然、申請者（コアリーダーJ）が、将来、放医研を担うようなリーダーとしての素質を期待されているものと理解している。その観点から、提案内容は自分のものとなっているか、単に外国の研究者に教えを請うのではなく、自分が研究を推し進めるという気概に満ちているか、そのためにチームの状態を把握、統御できる素質があるか、なども重要であると考えている。その観点から、最終的な結論を出すためには、ヒアリングの実施が不可欠であるとの意見の一致をみた。

当初の事務局の説明では、「3 コアの採択を目途とする」ということであったが、審査の結果、本委員会としては 4 コアを採択することを結論とした。

4. 採択結果

本事業の趣旨に照らして、以下の 4 コア（課題）が採択に値するとの結論を得た。

コア番号	コア名	コアリーダーJ	コアリーダーF
15FL-A02	Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam	KONISHI, Teruaki（小西輝昭）	WANG, Jun
15FL-A05	Radiation damage mechanism at molecular level approached with physichemical technologies	KODAIRA, Satoshi（小平聡）	BARILLON, Remi
15FL-A07	Impact of heavy ion irradiation on stem cells and tissue regeneration	SHIMOKAWA, Takashi（下川卓志）	KASPER, Maria
15FL-A08	Whole gamma imaging	YAMAYA, Taiga（山谷泰賀）	PARODI, Katia

5. その他

4 月 30 日開催のヒアリングでは 4 名の申請者が研究テーマについて口頭で紹介したが、いずれもわかりやすく、核心をついた発表で、評価者に強い印象を与えるものであった。

しかし、ヒアリングにおける発表と申請書の内容との乖離が大きく、申請書の作成能力に関する疑問の意見が出された。本事業に限らず外部資金の応募などに使われる申請書においては、内容はもちろん重要であるが、同時に自分の計画や能力を如何にわかりやすくアピールするかが重要である。その観点から、今回の申請書には不明瞭な文章構成や、アピール度が物足りないと感じられるものが複数見受けられた。いずれの申請書も英文の作成に問題があるとは思えない状況であったので、英語、日本語にか

かわらず、この種の申請書の作成能力（研究能力ではなく）の問題ではないかとの指摘である。この種の問題の解決には、経験を積むことが不可欠と思われるので、今後、放医研の若手研究者が様々な機会を捉えてその能力を磨くことを望むものである。

また、本委員会としては、次の 2 点を申し上げておく。

１．　前章で述べたように、本事業の審査にあたっては、事業の趣旨に則り、新規性やリーダーとしての資質も重要視している。従って、今回採択されなかった課題が、全てサイエンスのレベルが低いと判断したわけではない事を申し上げる。

２．　上に述べたように、放医研の若手研究者が、申請書作成の技術において不利益をこうむっているのではないかと、強く危惧する場面があった。今後、研究現場において外部資金の獲得が重要になることは否めないので、若手研究者に申請の機会を多く与えるとともに、申請書の作成能力を向上させるような方策を検討されてはいかがかと思う。

以上、

第 3 期「放医研国際オープンラボラトリー」2015 年度活動に関する審査結果報告

放射線医学総合研究所長殿、
2016 年 6 月 29 日

今般、事務局の依頼を受けて、第 3 期「放医研国際オープンラボラトリー」（以下、本事業と呼ぶ）の 2015 年度活動に関する評価を実施したので、その結果を報告する。この結論は、委員 5 人の総意である。

1. IOL 委員会構成

IOL 選考・評価委員会（以下、本委員会と呼ぶ）は以下のメンバーから構成される。

IOL 選考・評価委員会（順不同）

- 簱野嘉彦（東京工業大学名誉教授）
- 丹羽太貫（福島県立医科大学特命教授）
- 榮　武二（筑波大学教授）
- 河野俊之（東京工業大学教授）
- 柴田裕実（大阪大学特任研究員、委員長）

事務局

村上健、福田茂一、藤田敬、伊藤悦子
（放射線医学総合研究所）

2. 募集、及び採択の経緯と結果（この部分、事務局記入）

本事業に関するこれまでの経緯を簡単にまとめる。

【募集】

- 2014 年 12 月 15 日：理事長決定で本事業を立ち上げ
- 12 月 25 日～2015 年 2 月 20 日：公募（応募課題数 8 課題）

【審査】

- 2 月 26 日～3 月 19 日：書類審査、メールによる意見交換。
- 3 月 20 日：委員 5 人及び事務局で、1 回目の IOL 選考委員会を開催。
- 3 月 21 日～4 月 6 日：再度の書類審査とヒアリングを実施すべき 4 課題の決定。
- 4 月 30 日：放医研においてヒアリング実施。引き続き、2 回目の IOL 選考委員会を開催。採択案を決定。

【審査結果】以下の 4 コア（課題）を採択。

コア番号	コア名	コアリーダーJ	コアリーダーF
15FL-A02	Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam	KONISHI, Teruaki（小西輝昭）	WANG, Jun

15FL-A05	Radiation damage mechanism at molecular level approached with physichemical technologies	KODAIRA, Satoshi （小平聡）	BARILLON, Remi
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15FL-A08	Whole gamma imaging	YAMAYA, Taiga （山谷泰賀）	PARODI, Katia

【審査結果報告】審査の経緯、結果に関して本委員会より放医研理事会に報告を行い、その後、コアリーダーへ通知を行った。

- 5 月 8 日：理事会宛の審査結果報告書を提出。
- 5 月 14 日：第 91 回運営連絡会議で報告
- 5 月 19 日：第 94 回理事会議で報告。
- その後、コアリーダーへの通知

【2015 年度の活動の評価】2015 年 6 月からの活動について、報告書及び口頭での報告を受け、その後質疑応答を行った。

- 2016 年 6 月 14 日：委員へ報告書送付。
- 6 月 28 日：IOL 委員会に対する口頭発表と質疑応答を実施。

3. 評価の観点

本委員会としては、本事業の趣旨は、以下のような点にその真髄があると理解している。

✓ 海外交流の促進を通じて、放射線に関する様々な分野における「高度な研究シーズの創出」を推進する。
✓ 海外のアクティブな研究者との交流を通じ、次期放医研の中核を担いうる革新的な研究テーマを創出する。
✓ 失敗を恐れない自由闊達な研究の着想を積極的に支援する。

従って、結果が出たかどうかではなく、どのようなチャレンジをしたか、またそれらが論理的な帰結であるのかどうかを重視した。単に外国の研究者に教えを請うのではなく、自分が研究を推し進めるという気概に満ちた行動をとったかどうかが重要であると考えている。

評価結果は、次の 3 つの結論、方向性に分類した。

1. 上記の観点から見て著しく劣るので、次年度への継続は不可、
2. このまま、継続するのが妥当、
3. 非常にうまく進んでおり、別途、独立したテーマにした方が良い。この場合、「研究シーズの創出」と言う観点から IOL からは卒業とする、

4.　各コアの評価結果

2015 年度の 4 コア活動について、報告書及び口頭での説明と質疑応答を行い、その後、委員の間で意見交換を行った。

2015 年度といっても，採択が決まったあと実質的には 7 月くらいからの活動であろう。それを考慮すると，全体的な評価としては，各コアともそれなりの交流実績，研究活動の方向性，初期段階の研究成果を出していると考えられる。従って、IOL 委員会としての結論は、

4 コアとも 2 年目への継続が妥当であると考える。

しかしながら、全コアに対し、更なる高みを目指していただくために、委員会としてはあえて次のような苦言を呈する。

真にグローバルな視点に立って、自己の研究プログラムの新規性・独自性を論じる力が欠けているようである。その中での自己の研究プログラムがどのような位置にあるかについて明示することが必要であるとともに、自己の研究プログラムによって新たに明らかにされる事実が放射線科学、さらにより広く科学・技術のどのような部分に位置付けられるかについての論述が希薄であるように感じる。

なお、各コアへのコメントも多数出されており、このレポートの最後に付属資料としてまとめておく。

5.　IOL の事業全体に対するコメント

一般に、研究そのものと研究者育成の活性化のためには研究の国際化または研究環境の国際化が不可欠であり、科学技術基本法にも述べられている基本的な理念である。基礎研究および基盤研究については、その重要性はさらに大きいと思われ、すでに多くの大学等では外部資金申請のための萌芽的研究計画の活性化が行われている。しかしながら機構や研究所の多くは、国による潤沢な研究経費のサポートが永く続いたことによって、研究者自身がこのような状況の大きな変化に追随できない状況が残されている。

このような状況のもと、第 3 期 IOL は「放射線医学・生物・物理・化学・工学等戦略的に重要な研究分野において、海外のトップレベルの研究者の支援の下に、若手研究者が国際レベルの先端的な研究等を行う環境を整備することにより、云々」とされており、中堅や若手の研究者のレベルアップ、あるいは意識改革を促し、世界的な放射線研究の振興を目指すことに合致したシステムと考えられる。

IOL の予算が研究費を含んでおらず、海外交流のための旅費に限定されていることで、かえってコアメンバーが科研費などの外部資金に積極的に応募する動機付けになっている。外部資金を獲得できた、できないに関わらず、この方針も各コアリーダーにはよく理解されており、海外の交流相手先でも積極的な資金獲得に努力しているようである。また外部資金の申請において、新たな研究シーズを狙っていることに加えて、所属機関がこのようなサポートを継続的に行っているということは審査時の評価ポイントになっている可能性もある。このような点からも IOL が外部資金獲得のための研究の準備段階において重要な潤滑油としての役割を果たしていると確信している。

海外交流、研究ネットワーク構築に関して、放医研側の各グループは関連する海外の研究グループとこれまでにも個別に交流はあったであろうが、本 IOL のように「相手先」と「期間」を明確に決めて「新しい研究テーマの創出」を目的にプロジェクトを実施することで、単なる意見交換や情報交換ではなく、最終目標とそこまでのスケジュールの構築、目標達成のための役割分担等を意

識した研究活動をしなければならず、その結果、より実りの多い成果を期待することができる。本 IOL は、世界規模で研究者間の協力体制を、研究活動の積み上げによって築くものであり、国際的研究活動を推進する手法として、また、将来の研究テーマと人材を国際的規模で育てる活動としてユニークであり、是非とも継続して実行し、より一層の充実を期待する。さらには、産官学連携や異分野交流といった方面でも同様の取り組みができれば、今までとは違った研究成果も期待できる。

なお、委員の一人より、次のようなコメントが寄せられていることを申し添える。

【IOL の必要性】

1954 年のビキニ海域における漁夫の被ばくをきっかけに国論が沸騰し、1957 年には、放射線の健康リスクの実態と機構を明らかにする研究所として放医研が創立された。それから 60 年になろうとする昨今、放射線影響研究は世界的にも研究者人口が減少し、その数は今やクリティカルマスを切った感がある。この状況のもと、力強い研究を行う放医研を目指すには、まず世界の研究者を糾合して研究にあたる体制が重要である。国際オープンラボは「放射線医学・生物・物理・化学・工学等戦略的に重要な研究分野において、海外のトップレベルの研究者の支援の下に、若手研究者が国際レベルの先端的な研究等を行う環境を整備することにより、ひいては研究所全体のレベルアップを図るため、設置する」とされており、世界的な放射線研究の凋落に歯止めをかけ、先端研究としての放射線影響研究を目指すには、恰好のシステムである。そのため、現段階では個々の申請者の人脈で拠点形成をしているが、IOL 全体としてたとえば欧州の MELODI のような組織と結ぶ努力は、考慮に値するものと考える。

以上、

【付属資料】各コアの活動報告へのコメント

15FL-A02 （小西） 評価:2 年目への継続を推奨	● 本申請者は世界的にみても技術的に高い水準にあるプロトンマイクロビームを開発し、それを駆使して細胞に対する放射線損傷の基本過程を研究しており、しっかりした考えの下で研究計画を建て、研究を進めているようである。 ● マイクロビーム装置は研究のための道具であるからには、それを使ってどのような研究を行うかがカギになる。申請者はバイスタンダー効果など、世界的にもよく知られている現象について本装置を使って解析しようとしているのは、一つの在り方であろう。しかしせっかくの高性能の装置であるのなら、たとえば細胞質と核、それに両方の３つの照射状況のもとで遺伝子発現パターンがどのように異なるかといったミクロの状況から、部分照射が臓器全体の機能にどのような影響を与えるのかといったマクロな臨床までつなぐ努力を行うことで、より新しい展開が期待できる。研究の方向の面での工夫が欲しい。 ● マイクロビームでなくてはできない研究に絞り，さらには新規性を狙うという進め方はよい。申請者に明確なコンセプトがありそうに見え，活動報告もわかりやすかった。相手先機関も含め外部資金の獲得にも成功している。本事業への応募当初から，新たな研究テーマの創出に加え，割とネットワーク構築を強調し，マイクロビームトレーニングコースの設置とアジアにおけるセンター化を目指しているようだが，その方面の取り組みにおいても努力している様子がうかがえ，特に海外交流の面で今後が期待される。ただ、その目標が国内初またはアジア初に閉じているように見える。よりグローバルな視点が欠けているようである。 ● マイクロビームといっても性能や使い勝手がまちまちであるので、各国、各機関の装置の性能を生かした研究手法が求められ、加速器利用研究においてはそこにこそ、このような研究拠点のネットワークを構築する意味があると思われるので、そのような方向での発展を望む。 ● マイクロビームを用いた放射線誘発細胞防御応答シグナルの解析をテーマとしているが、SPICE-NIRS マイクロビームの他のマイクロビームに比べた特徴に関する論述、つまりこのテーマについての特徴的な新しい成果に関する主張が乏しいように感じられる。 ● それぞれの施設の特徴を生かしたネットワーク構築を期待し、継続して研究を進めて頂きたい。

15FL-A05 （小平） 評価:2 年目への継続を推奨	● 申請者は「加速重イオンビーム物理学」の立場と経験から、物質（多分人体も含まれる）に対する放射線損傷のメカニズムを分子レベルで解明しようと述べている。委員の一人は放射線と物質の相互作用の物理化学・化学物理学を専門としている立場から、そして、この相互作用の物理学（放射線作用初期過程の解明）、それに続く放射線効果の物理学、化学、生物学等に関する研究の評価・検討・展望を行った長期にわたる国際プロジェクトを主宰した立場から、本研究は大変貴重なアプローチであると結論している。 ● 具体的な問題として、物理プロセスと化学プロセスを繋げる物理量が LET あるいは dose によって表されているが十分な検討が必要と思われる。これまでも色々なアプローチがされているが、まだ的確なものは見当たらないので、難しいかもしれないが、可能性のあるアイデアが示されると素晴らしい研究になる。報告で示されたカーボンのトラックはカーボンのみに依るのか、フラグメントの影響はないのか、との疑問もあるので、トラック内粒子の弁別が重要であると考えられる。 ● ネットワークにおける活動報告では，各グループ間の役割分担がよくわからなかった。もちろん，必ずしもグループごとに異なった役割をもたないといけないというわけではないが，それぞれのグループが実績をもっている分野を組み合わせれば，より強力なチームになると思われるので，そのあたりの説明があるとわかりやすい。照射損傷において上記グループ同様，新たなメンバーの追加，また唯一国内他機関のメンバーが入っているなど，交流面では実績をあげている。 ● 一般に、ある分野の研究に対して今までとは大きく異なる分野からのアプローチを行うことによって、新規性・独創性に富んだ新しい研究成果を生むことにつながる可能性が大きくなると言われることが多い。その場合に、アプローチの対象となる分野ですでに蓄積されている知見、成果等の情報をあまり豊富に把握していない方がよいとも言われている。しかし、「新規性・独創性に富んだ研究成果」を得るためには、対象分野との真の研究交流（意見・コメント等を戦わせること）が必須であるので、大いなる努力を望む。 ● 重要な課題を是非とも解決してもらいたいので、継続して研究を進めて頂きたい。

15FL-A07 （下川） 評価:2 年目への継続を推奨	● 本申請では重粒子線による組織傷害とその治癒に関する特異性を明らかにすること、つまり「キズとは何か？」から説き起こして、研究成果の「科学」（これは、「放射線の科学」および「いわゆる科学全体」を指すのだろうか？）への展開を明らかにしたいと述べている。 ● 大変貴重な問題提起であると考えられるが、研究テーマにある重イオン照射による「キズ」とその他の放射線（X 線、電子線等）による「キズ」を識別する方法とその「科学」への繋がりが何であるか、また、それを明らかにする戦略がまだ明確でないように思われる。 ● 用いている実験系は、申請者は扁形動物のプラナリア、海外研究者は高等動物である哺乳類のマウスで、相互に異なっている。これら２つのシステムを用いて傷の治癒の研究を行い、何らかの結論を出すためには、申請者と海外の研究者が考え方やデータを共有する必要がある。まったく異なる実験動物であっても共有でききるのは傷の治癒に関わるシグナル系とそれに関わるサイトカイン等の分子レベルの因子であろう。しかしながら本申請では分子経路の解析が無いように思われ、これからの先行きが気にかかる。 ● プラナリアの解析手法に関し、MRI や PIXE の利用など解析方法にもある程度成果があったようであるが、まだ解析の方向性が見えていないことは問題である。3D プリンティングの応用についてはテーマが更に拡がる可能性があると思われ今後に期待できる。 ● ネットワークに関しては交流相手先グループとは Skype meeting も含めてコミュニケーションをよく取っているようで、共同で HIMAC での照射実験も行ったとのこと、初年度としては予定通りの進捗状況であろうか。グループ内での議論により途中から新たなグループを加えたことは、研究面だけでなく海外交流の面からも望ましい結果である。残念ながら科研費は獲得できなかったようだが，相手先も含め，今後も積極的にトライしてほしい。 ● 研究の方向性を迅速に明確にして、継続して頂きたい。

15FL-A08 （山谷） 評価:2 年目への継続を推奨	● 申請者は放射線を利用したイメージング技術の開発に多くの実績を持っており、本研究は従来の PET 技術に新たな視点を導入しようとする試みで、放医研における PET 技術のレベルの高さを示すものである。研究の新規性が豊かであり、研究目的・目標を明確に定めた上で、本 IOL を有効に活用して研究計画を立て、推進している点は評価に値する。 ● 本 IOL で期待している研究ネットワーク構築について、交流相手先は LMU のみであるが，検出器開発に関してはお互いに実績があり，その得意分野を融合して新しいシステムを開発するという方向性は妥当で，相手先との連携はうまくいっているように見える。また、本 IOL が科研費等の外部研究資金の獲得とそれによる研究の活性化を行うための潤滑油になっているようであり、既に外部資金の獲得にも成功しており、今後の成果が期待される。その反面、申請者の本研究の進め方は本 IOL が期待している冒険的な研究でなく、またはその結果として、予期に反する新しい成果を獲得する機会に出会う可能性が少ないのではないかと推測される。 ● 技術的な面からは PET＋Compton 検出器で感度を上げることは理解できるが、さすがに応募書類にあったように 1 event というわけにはいかないだろうから、PET 単独に比べてどのくらい優位性があるのか、シミュレーションで明らかにして頂きたい。低レベルの分析が被ばくの低減だけで無く、どのような医学的な機能、応用に繋がるのかを分かりやすく示されると良いであろう。 ● ベクレル・オーダーのイメージングを達成するこの研究開発が成功すれば、PET 診断に付随する線量が大幅に低減されることになり、医療におけるインパクトは計り知れないと考えられるので、早期実現を目指して研究を進めて頂きたい。

第 3 期「放医研国際オープンラボラトリー」2016-2017 年活動に関する評価結果

放射線医学総合研究所長殿、
2017 年 7 月 6 日

今般、事務局の依頼を受けて、第 3 期「放医研国際オープンラボラトリー」(以下、本事業と呼ぶ)の 2016-2017 年度活動に関する評価を実施したので、その結果を報告する。この結論は、委員 5 人の総意である。

1. IOL 委員会構成

IOL 選考・評価委員会（以下、本委員会と呼ぶ）は以下のメンバーから構成される。

IOL 選考・評価委員会（順不同）

- 簇野嘉彦（東京工業大学名誉教授）
- 丹羽太貫（福島県立医科大学特命教授）
- 榮　武二（筑波大学教授）
- 河野俊之（東京工業大学教授）
- 柴田裕実（大阪大学特任研究員、委員長）

事務局

村上健、福田茂一、松藤成弘、藤田敬、伊藤悦子、
（放射線医学総合研究所）

2. 主な活動経緯（この部分、事務局記入）

本事業に関する 2016-2017 年の経緯をまとめる。

【審査対象コア】以下の 4 コア

コア番号	コア名	コアリーダーJ	コアリーダーF
15FL-A02	Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam	KONISHI, Teruaki (小西輝昭)	WANG, Jun
15FL-A05	Radiation damage mechanism at molecular level approached with physicochemical technologies	KODAIRA, Satoshi (小平聡)	BARILLON, Remi
15FL-A07	Impact of heavy ion irradiation on stem cells and tissue regeneration	SHIMOKAWA, Takashi (下川卓志)	KASPER, Maria

15FL-A08	Whole gamma imaging	YAMAYA, Taiga （山谷泰賀）	PARODI, Katia

【2016-2017 年の主な活動と出来事】

- 2016 年 4 月 1 日：QST 発足
- 2016 年 6 月 28 日：2015 年度の活動について、IOL 委員会に対する口頭発表と質疑応答を実施。
- 2016 年 6 月 29 日：IOL 委員会報告書作成（提出済み）
- 2016 年 7 月：IOL 活動に関して所長説明（昨年度分でここまで報告済み）

- 2016 年 10 月：仏ストラスブール大学より来日、研究打ち合わせと HIMAC 実験（A05）
- 2016 年 11 月：イタリア出張（A07）
- 2016 年 11 月：放医研での実験の為、外国人研究者招聘（A02）
- 2017 年 1 月：ドイツ出張（A08）
- 2017 年 2 月 28 日：IOL シンポジウム 2017
- 2017 年 5 月 23 日：IOL 委員に評価用資料送付
- 2017 年 6 月 8 日：IOL 委員からの評価書締切

その他に、月一回の定期打合せ（事務局、及びコアリーダー参加）

【制度に関する変更点】

QST 発足に伴う組織の変更などから、IOL の活動にもある期間、活動の停滞が避けられなかった。また、外国とのやり取りが主になる場合、4 月 - 翌年 3 月の活動期間はあまり好ましいものでは無い事が実感され、9 月-翌年 7 月に再定義することを事務局が提案し、委員の賛同を得た。

3. 評価の観点

本委員会としては、本事業の趣旨は、以下のような点にその真髄があると理解している。

✓ 海外交流の促進を通じて、放射線に関する様々な分野における「高度な研究シーズの創出」を推進する。
✓ 海外のアクティブな研究者との交流を通じ、次期放医研の中核を担いうる革新的な研究テーマを創出する。
✓ 失敗を恐れない自由闊達な研究の着想を積極的に支援する。

従って、結果が出たかどうかではなく、どのようなチャレンジをしたか、またそれらが論理的な帰結であるのかどうかを重視した。単に外国の研究者に教えを請うのではなく、自分が研究を

推し進めるという気概に満ちた行動をとったかどうかが重要であると考えている。

4.　コアの評価結果

2016-2017 年の 4 コア活動について、IOL シンポジウム 2017 の発表、及び報告書に基づき、各委員が意見をまとめ、それらを事務局でまとめて委員に送り最終的な承認を得た。

第 3 期の活動予定を 3 年と設計していることも考慮し、IOL 委員会としての結論は、4 コアとも 3 年目への継続が妥当であると考える。但し、委員会としては、全てのコアの活動結果に満足しているわけではない。また、2 年目になり、コア間の活動状況に差が出てきたことも事実であり、今後の運営、あるいは次期 IOL の設計に関して心すべき点と思われる。全体に共通する問題点を次に列挙する。

- まず全体的に、統合に伴う混乱期にあったにも関わらず、活動が続いた点は評価する。
- 今季に限ったことではないが、報告書など書類の書き方や、説明の仕方が全体的に稚拙であるとの印象を受ける。外部資金獲得などの点からも必要な技術であり、事務局等を中心にした、組織的な教育、指導が必要と思われる。
- 研究の目的の明確化や、その研究が全体の中でどのような位置づけになるか、と言う認識が必ずしも十分ではない。この結果、目的達成のための戦略が不明確になっている。この改善のためには普段の議論、批判等が欠かせず、この点も事務局による改善の努力が必要である。

5.　IOL の事業及び運営に関するコメント

第 3 期 IOL は「放射線医学・生物・物理・化学・工学等戦略的に重要な研究分野において、海外のトップレベルの研究者の支援の下に、若手研究者が国際レベルの先端的な研究等を行う環境を整備することにより、云々」とされており、中堅や若手の研究者のレベルアップ、あるいは意識改革を促し、世界的な放射線研究の振興を目指すことに合致したシステムと考えられる。研究活動の国際化によって研究環境の活性化をもたらし、その結果として、新規性に富んだ高度の研究成果を得ることにあると考えられる。以下が事務局、及び運営に対する委員の意見である。

（1）限られた諸条件の下で、その実現と継続のために、大きな熱意と工夫に基づいた運営が行われていることは高く評価され、また今後に期待されるところは極めて大きい。特に 2016-2017 年は新機構発足による混乱により、有形無形の影響があったであろうが、事務局の努力により影響は最小限に抑えられたと考えられ、その点は評価する。

（2）しかしながら、計画の立案、報告書の書き方などには、様々な問題点が含まれており、第 3 期 IOL の理念を十分に反映したものとは言えない。コアリーダーの努力だけではなく、レベルアップを図るシステムを事務局がリードして作製することを強く希望するものである。

（3）IOL の予算が研究費を含んでおらず、海外交流のための旅費に限定されていることで、かえってコアメンバーが科研費などの外部資金に積極的に応募する動機付けになっている。外部資金を獲得できた、できないに関わらず、この方針も各コアリーダーにはよく理解されており、海外の交流相手先でも積極的な資金獲得に努力しているようである。

海外交流、研究ネットワーク構築に関して、放医研側の各グループは関連する海外の研究グループとこれまでにも個別に交流はあったであろうが、本 IOL のように「相手先」と「期間」を明確に決めて「新しい研究テーマの創出」を目的にプロジェクトを実施することで、単なる意見交換や情報交換ではなく、最終目標とそこまでのスケジュールの構築、目標達成のための役割分担等を意識した研究活動をすることになり、その結果、より実りの多い成果を期待することができる。

本 IOL は、世界規模で研究者間の協力体制を、研究活動の積み上げによって築くものであり、国際的研究活動を推進する手法として、また、将来の研究テーマと人材を国際的規模で育てる活動としてユニークであり、是非とも継続して実行し、より一層の充実を期待する。さらには、産官学連携や異分野交流といった方面でも同様の取り組みができれば、今までとは違った研究成果も期待できる。

以上、

2018 年 7 月 24 日

第 3 期「放医研国際オープンラボラトリー」活動に関する評価結果

放射線医学総合研究所長殿

今般、事務局の依頼を受けて、第 3 期「放医研国際オープンラボラトリー」（以下、本事業と呼ぶ）の活動に関する評価を実施したので、その結果を報告する。この結論は、委員 5 人の総意である。なお、今回の評価は本事業全体に対するものであり、2015 年 6 月～2018 年 7 月に渡る活動を対象とする。

1. IOL 委員会構成

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- 丹羽太貫（放射線影響研究所理事長）
- 榮　武二（筑波大学教授）
- 河野俊之（東京工業大学名誉教授）
- 柴田裕実（大阪大学特任研究員、委員長）

事務局

村上健、福田茂一、松藤成弘、藤田敬、伊藤悦子、水落佳子
（放射線医学総合研究所）

2. 本事業発足の経緯（この部分、事務局記入）

本事業は以下のようなスケジュールで開始された。

- 2014 年 12 月 15 日：理事長決定で本事業を立ち上げ
- 同 12 月 25 日：公募開始
- 2015 年 2 月 20 日：公募締め切り、8 課題応募有
- 同 3 月 20 日：委員 5 人及び事務局が東京八重洲ホールで、1 回目の IOL 選考委員会を開催。
- 同 4 月 30 日：放医研においてヒアリングを実施。引き続き、2 回目の IOL 選考委員会を開催。採択案を決定。

その結果、応募 8 課題から以下の 4 コアが採択された

コア番号	コア名	コアリーダーJ	コアリーダーF
15FL-A02	Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam	KONISHI, Teruaki（小西輝昭）	WANG, Jun
15FL-A05	Radiation damage mechanism at molecular level approached with physicochemical technologies	KODAIRA, Satoshi（小平聡）	BARILLON, Remi

15FL-A07	Impact of heavy ion irradiation on stem cells and tissue regeneration	SHIMOKAWA, Takashi （下川卓志）	KASPER, Maria
15FL-A08	Whole gamma imaging	YAMAYA, Taiga （山谷泰賀）	PARODI, Katia

審査の経緯、結果に関して本委員会より放医研理事会に報告を行い、その後、コアリーダーへ通知を行った。

- 2015 年 5 月 8 日：理事会宛の審査結果報告書を提出。
- 同 5 月 14 日：第 91 回運営連絡会議で報告
- 同 5 月 19 日：第 94 回理事会議で報告。
- その後、コアリーダーへの通知、活動を開始

3. 主な活動経緯（この部分、事務局記入）

各コアの活動の詳細は、別途、活動報告が作成されているのでそちらを参照の事

【2015 - 2018 年の主な活動と出来事】

- 2015 年 6 月：各コアが活動開始
  - 運営交付金より各コア 300 万円の予算措置がなされた
  - 2016 年 4 月 1 日：QST 発足
- 2016 年 6 月 14 日：各コアより 2015-2016 年の活動報告書を IOL 委員会に提出
- 同 6 月 28 日：IOL 委員会に対する口頭発表と質疑応答を実施
- 同 7 月：IOL 活動に関する評価報告書を放医研所長に提出、並びに所長説明を実施
  - 同 7 月：寄附金より各コア 100 万円の予算措置が取られた。3 年目も同様。
- 2017 年 2 月 28 日：IOL シンポジウム 2017
- 同 5 月：2016-2017 年の活動報告書を IOL 委員に提出
- 同 7 月：IOL 活動に関する評価報告書を放医研所長に提出
- 2018 年 6 月：各コアより 2015-2018 年の活動報告書を IOL 委員に提出
- 2018 年 6 月 15 日 IOL シンポジウム 2018
- 同 7 月：今回の報告書提出

◎ その他に、月一回の定期打合せ（事務局、及びコアリーダーが参加）

【制度に関する変更点】

本事業の開始は、様々な事情により 2015 年 4 月ではなく 6 月にずれ込んだ。また、外国とのやり取りが主になる場合、4 月 - 翌年 3 月の活動期間はあまり好ましいものでは無い事が、本事業発足後に実感された。更に、QST 発足に伴う組織の変更などから、IOL の活動にもある期間、活動の停滞が避けられなかった。これらの事情を考慮して、事業の途中で、本事業の一年の区切りを 9 月から翌年 7 月に再定義することを事務局が提案し、IOL 委員の賛同を得た。

4. 評価の観点

本委員会としては、本事業の趣旨は、以下のような点にその真髄があると理解している。

- ✓ 海外交流の促進を通じて、放射線に関する様々な分野における「高度な研究シーズの創出」を推進する。
- ✓ 海外のアクティブな研究者との交流を通じ、次期放医研の中核を担いうる革新的な研究テーマを創出する。
- ✓ 失敗を恐れない自由闊達な研究の着想を積極的に支援する。

従って、結果が出たかどうかではなく、どのようなチャレンジをしたか、またそれらが合理的帰結であるのかどうかを重視した。単に外国の研究者に教えを請うのではなく、自分が研究を推し進めるという気概に満ちた行動をとったかどうかが重要であると考えている。

5. コアの評価結果

2015 - 2018 年の 4 コアの活動について、過去の口頭発表、IOL シンポジウムでの発表、及び報告書に基づき、IOL 委員が各自の意見をまとめ、それらを事務局でまとめた。その詳細はつまびらかにはしないが、各コアリーダーには、当該コアに関する全委員の意見を通知した。

これまでも、本事業の制度設計に基づき、一年ごとに各コアの評価を行い、その後の事業継続の可否を判断した。結果的に 4 コアとも 3 年余りの活動が続けられたことは、コアリーダーの努力によるものと評価している。

1 回目、2 回目の評価においては、次のような問題点が指摘された。

- 報告書など書類の書き方や、説明の仕方が全体的に稚拙であるとの印象を受ける。外部資金獲得などの点からも必要な技術であり、事務局等を中心にした、組織的な教育、指導が必要と思われる。
- 研究の目的の明確化や、その研究が全体の中でどのような位置づけになるか、と言う認識が必ずしも十分ではない。この結果、目的達成のための戦略が不明確になっている。この改善のためには普段の議論、批判等が欠かせず、この点も事務局による改善の努力が必要である。

これらの指摘に関しては、その後、コアリーダーと事務局の努力により、かなりの改善が見られたものと評価する。本事業終了時（つまり、今回の評価）の評価の過程で出された、IOL 委員から寄せられたコメントの一部を紹介する。

- 交流においても海外のグループと対等、あるいはそれ以上のイニシャティブを取って共同研究を推進したと考えられ、これも特筆に値する。
- オープンラボに関与したコアリーダーをはじめとする研究者は、各自の研究の成果や進捗に支えられ、リーダとしてまた研究者としての自信を得たように感じている。そして成果に支えられた新たな研究への挑戦は、本国際オープンラボが量子科技研開発機構もたらした最大の貢献である。
- 前回、指摘した事項は、多くのグループで大幅に改善されつつあるようである。

以上の事から、4 コアとも本事業の趣旨を十分に理解し、様々な問題を克服して、当初目指した目的を十分に達成したものと評価する。

6.　IOL の事業及び運営に関するコメント

第 3 期 IOL は「放射線医学・生物・物理・化学・工学等戦略的に重要な研究分野において、海外のトップレベルの研究者の支援の下に、若手研究者が国際レベルの先端的な研究等を行う環境を整備することにより、云々」とされており、中堅や若手の研究者のレベルアップ、あるいは意識改革を促し、世界的な放射線研究の振興を目指すことに合致したシステムと考えられる。研究活動の国際化によって研究環境の活性化をもたらし、その結果として、新規性に富んだ高度の研究を推進することにあると考えられる。以下が事務局、及び運営に対する委員の意見である。

（1）限られた諸条件（予算、人的資源）の下で、その実現と継続のために、大きな熱意と工夫に基づいた運営が行われていることは高く評価され、また今後に期待されるところは極めて大きい。特に 2016 年の新機構発足による混乱により、有形無形の影響があったであろうが、事務局の努力により影響は最小限に抑えられたと考えられ、その点は高く評価する。

（2）本事業発足当初は、各コアの計画の立案、報告書の書き方などに、様々な問題点が含まれており、本事業の理念を十分に反映したものとは言えなかった。しかし、終了時には、「IOL の理念」は、ほとんどすべてのコア・グループについてほぼ達成されているか、または達成しつつあることが確認された。コアリーダーの努力だけではなく、事務局がリードしてレベルアップを図ってきたことの表れと考えられる。それは単なる事後チェック的なスタンスではなく，各コアの活動に事務局が積極的に関与する、つまり良い意味で、事務局が研究活動（研究内容ではなく）に関与することの成功例になったのではないか。このことは、事務局は単なる事務取扱いでは無く、高度の研究レベルを備えた人材の集団である必要性を示している。

（3）IOL の予算が研究費を含んでおらず、海外交流のための旅費に限定されていることで、かえってコアメンバーが科研費などの外部資金に積極的に応募する動機付けになっている。外部資金を獲得できた、できないに関わらず、この方針も各コアリーダーにはよく理解されており、海外の交流相手先でも積極的な資金獲得に努力している点は評価できる。人的交流のための「旅費・滞在費」の確保が困難である状況に置かれている中で「新しい IOL の理念」のもとで、明らかに、よく工夫され熱意のある運営の結果として注目すべき成果を実現したことは高く評価されると結論したい。

本 IOL は、世界規模で研究者間の協力体制を、研究活動の積み上げによって築くものであり、国際的研究活動を推進する手法として、また、将来の研究テーマと人材を国際的規模で育てる活動としてユニークであり、是非とも継続して実行し、より一層の充実を図る事を期待する。さらには、産官学連携や異分野交流といった方面でも同様の取り組みができれば、今までとは違った研究成果も期待できる。

以上

２．第３期 IOL 各コアの活動報告書

SPICE-BIO research core

Studies on radiation induced defensive cellular signaling using SPICE-NIRS microbeam

CORE LEADER (J)

Teruaki Konishi / Ph.D Senior Researcher
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Expertise:
radiation biology / microbeam / heavy ions/ bystander response / DNA double strand breaks

CORE LEADER (F)

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Expertise:
radiation biology / oxidative stress response / mitochondrial biogenesis

Core Members

- Dr. Narongchai Autsavapromporn, Ms. Nahathai Dukaew,
  Dr. Ariyaphong Wongnoppavich Dr. Wanisa Punfa,
  (Faculty of Medicine, Chiang Mai University, Thailand)
- Dr. Tengku Ahbrizal Farizal Tengku Ahmad, Dr. Hashim Asmaliza
  (Malaysian Nuclear Agency, Malaysia)
- Prof. Li-jun Wu (Hefei Institute of Physical Science, Chinese Academy of Sciences, China PR)
- Dr. Lu DONG (Institute of Modern Physics, Chinese Academy of Sciences, China PR)
- Dr. Gen Yang (Department of Physics, Peking University, China PR)
- Mr. Sebastian Deigeler, Dr. Luis Spitta
  (DLR German Aerospace Center, Germany)

- Ms. Alisa Kobayashi, Mr. Masakazu Oikawa,
  Dr. Daisuke Ohsawa, Dr. Chuiha Liu, Dr. Yoshiya Furusawa
  (National Institute of Radiological Sciences, QST)

Purpose of the Project:

Classical radiation biology asserts that the initial damage induced by the energy deposited directly into DNA during exposure to radiation is the main cause of the multitude of reported radiobiological consequences. Thus, DNA is considered to be the primary target of radiation. However, in the past several decades, many phenomena have been reported that cannot be explained by this classical dogma. These findings indicate the existence of non-DNA/secondary targets that may affect the fate of irradiated and nearby non-irradiated cells.

The non-DNA/secondary targets may be activated by signals received from non-irradiated cells through bi-directional inter-cellular communication. The radiation-induced bystander effect (RIBE) involves non-irradiated cells that exhibit irradiated effects as a result of signals received from nearby irradiated cells. Thus, these non-DNA/secondary targets in the irradiated cells may be directly activated by energy deposition in the cytoplasm or indirectly activated by the RIBE to propagate signals from the surrounding non-irradiated cells. Our aim is to identify non-DNA/secondary targets and examine their involvement in the damage incurred by the primary target (DNA) using microbeam technology. For the two specific aim,

Microbeam is a powerful tool that can target the cell nucleus and/or cytoplasm with few micrometers resolution. Using this technique, it is possible to distinguish the non-targeted cells from the targeted cells, which is essential in studying the radiation induced bystander effect. The microbeam irradiation system used in this study, Single-Particle Irradiation system to Cell (SPICE) at the National Institute of Radiological Sciences (NIRS), provides a 3.4 MeV proton microbeam focused with a quadrupole magnetic lens on an upward vertical beam line. At present, SPICE-NIRS microbeam is the only proton microbeam facility in Asia where a single-ion/single-cell irradiation can be performed on mammalian cells with enhanced stability and high throughput capacity, using an upward vertical beam with a diameter below two micrometers.

The goal of this research study was to investigate two specific aspects of the underlying mechanisms of secondary targets of radiation in cells. Firstly, we investigated the cellular response induced by cytoplasmic damage, focusing on the activation of the oxidative stress response pathway triggered by Nuclear factor-erythroid 2 related factor 2 (NRF2). This transcription factor is known to regulate cellular oxidative status by controlling the expression of many detoxifying enzymes, including those that participate in the maintenance of oxidative equilibrium in cells. In the second part of our study, we examined the bi-directional response resulting in the “rescue” of targeted cells, which has been described as a facet of the radiation-induced bystander effect. In addition, we determined the characteristics of radiation-induced cell signaling between two different types of cell lines, normal and cancer cells. In this part of the project, our aim was to characterize the bystander signaling molecules underlying the cellular response to radiation exposures.

The overall strategy of our research core is to establish an international collaborative network investigating radiation biology in Asia, using advanced microbeam technology to expand our studies of radiation-induced cellular signaling. To maintain the continuity of this collaborative network and to project its efforts beyond those of the NIRS-International Open Laboratory (IOL) program exclusively, we would like to establish a “microbeam training course” at the NIRS to recruit young investigators in Asia to participate in the study of microbeam biology.

The framework of the proposed studies was constructed and directed with the strong partnership between

Core Leader J, T Konishi and Core Leader F, Prof. Jun Wang (Hefei Institute of Physical Science, CAS), who are both researcher in the field of radiation biology with different expertise. TK is the main developer of the SPICE-NIRS microbeam and currently working on DNA double strand break repair and radiation induced bystander effect/inter-cellular communication. Prof. J Wang's expertise in oxidative stress cellular response, mitochondira biogenesis, and cytoplasm damage response and a person with the knowledge and experience would be the perfect counterpart for the studies on microbeam biology.

Summary of the Project

The aimed goals of SPICE-BIO research core and the achievements are summarized as follows:

1) ***Financially independent research structure for an international collaboration***: All the members put enormous effort in achieving external research fundings. Core leader J, TK was awarded for "JSPS Grant-in-Aid for Exploratory Research" in 2016 and "JSPS Grant-in-Aid for Scientific Research B" in 2017.and Core leader F, JW was also awarded for "Regular project of Natural Science Foundation Committee of China" in 2016 and was accepted for Chinese Scholarship Council program to stay in NIRS for 6 months in 2018. The core's activities were mostly supported by these grants. Other than these, A Kobayashi (NIRS-QST) was also accepted for "JSPS Grant-in-Aid for Young investigator's research B". Also international members were successful, for eg, N Autsavapromporn was accepted for multiple grants of Thailand, and also by JSPS for fellowship to perform experiment in NIRS. The high productivities of our research core results in five peer-review papers, and three papers submitted currently.

2) ***Constructing an international partnership among the members and the co-workers***: Among the members, we challenged for JSPS "Core to core program B" for constructing microbeam radiobiology platform in Asia. Concurrently, we tried for bi-lateral partnership program of Chinese Academy of Science, "Key project of International Partnership Program". Unfortunately we did not succeed. In the reviewers comments strongly suggested to have pre-arranged official agreements among the affiliations. To be prepared for our upcoming challenges, we made official agreements on the "Memorandum of cooperation between the NIRS and Faculty of Medicine, Chiang Mai University" in the Sept 2017, and also "Memorandum of cooperation between Columbia University and QST" in March 2018.

3) Establishing "microbeam training course" in NIRS: Representative model is the microbeam training course (MTC) held at RARAF, Columbia University. TK have contacted Dr. Marcelo Vasquez, director of the RARAF MTC from 2015 to seek a way for cooperation to hold a MTC in NIRS. We included the MTC program in the proposals (mentioned in section 2)), which we did not succeed. However, we were successful in constructing platform for younger investigators, Ph.D graduate to have their studies using microbeam technology under the platform of IOL. For example, semiars with JW's graduate students, NA's graduate joined our IOL research core and stayed in NIRS for over 5 months, AK enrolled in graduate school of Tsukuba University, and so on.

Altogether, the activities of SPICE-BIO research core strongly accelerated the scientific studies of individual members, and also strengthen the collaboration. Furthermore, each member expanded the research core's framework and recruited younger investigators to participate in our activities.

*A. **Defensive cellular response by cytoplasm irradiation***

A-1. Enhanced DNA double-strand break repair by sequential microbeam irradiation targeting the nucleus and cytoplasm of cells

The cell line used in this research was normal human lung fibroblast WI-38. Experiments were conducted on the SPICE-NIRS proton microbeam irradiation system. We examined the speed of DNA double-strand break (DSB) repair in microbeam-irradiated WI-38 normal human fibroblast cells that were targeted in the nucleus, cytoplasm, or both, using SPICE-NIRS microbeam technology. Cells were fixed at various time points between one and twenty-four hours post-irradiation. Subsequently, they were immunostained using antibodies against γ-H2AX, which is used as marker for DSB, to quantify the residual DSB per nucleus in images obtained using a microscope. Microbeam irradiation induced significant γ-H2AX activity in proportion to the number of protons delivered per nucleus. In nucleus-targeted cells, γ-H2AX levels did not increase significantly in comparison with non-irradiated controls one hour post-irradiation. However, at four hours post-irradiation, γ-H2AX levels were significantly higher than in the controls, and the increase was proportional to the number of protons delivered. For the cells irradiated with 500 protons per nucleus, we found less residual γ-H2AX with additional 200 and 500 proton deliveries to the cytoplasm at 16 hours and 24 hours post-irradiation, respectively. Taken together, cytoplasmic damage enhances repair of DSB induced by irradiation of the nucleus. Further study is needed to identify the type of cytoplasmic damage and mechanism of intracellular signaling responsible for the enhanced cellular response to DSB.

A-2 Cytoplasm irradiation induced Nrf2-mediated anti-oxidative pathways

Nuclear factor-erythroid 2 related factor 2 (Nrf2)-mediated antioxidative pathways play very important roles in maintaining the equilibrium of cellular oxidative status. Under normal or unstressed conditions, NRF2 is kept in the cytoplasm by association with a cluster of proteins that degrade it through the ubiquitin-mediated pathway. When cells are exposed to stress, NRF2 will dissociate from the protein complex and translocate from cytoplasm to nucleus where it binds DNA and initiates the transcription of its targets. Chemicals that stimulate NRF2 pathways can attenuate the cellular damage induced by various types of stresses. Based on previous findings that cytoplasmic irradiation resulted in nuclear DNA damage, in this IOL program term, we explored whether the NRF2-mediated antioxidative response was involved in cytoplasmic radiation-induced biological effects. Results are summarized as follows:

A-2-1 Activation of NRF2 alleviated cytoplasmic radiation-induced DSB

We confirmed that cytoplasmic irradiation stimulated the accumulation of NRF2 in the nucleus and the upregulation of its target gene, heme oxygenase-1 (HO-1). The level of NRF2 nuclear accumulation was dependent on the dose of cytoplasmic radiation applied, ranging from 0 to 1000 protons per cell. This response was enhanced with the elongation of post-irradiation time, up to 24 hours. Induction of NRF2 in the nucleus by 15 μM tert-butylhydroquinone (tBHQ) attenuated the levels of DNA double-strand breaks induced by cytoplasmic irradiation with 200 and 500 protons, which correlated with previous findings that XRCC4 and RAD51 levels in the nucleus of the irradiated cells were higher when cells were pre-treated with tBHQ.

A-2-2 Cytoplasmic irradiation resulted in mitochondrial fragmentation and down-regulation of

p53

Mitochondrial morphology in WI-38 cells was detected after exposure to cytoplasmic irradiation, revealing an increase in fragmented mitochondria. Inhibition of mitochondrial fragmentation with 50 μM Mdivi-1 suppressed cytoplasmic radiation-induced NRF2 nuclear accumulation and aggravated DSB level. We also observed that cytoplasmic irradiation down-regulated p53, which is usually up-regulated by whole-cell irradiation. Furthermore, we identified that p53 inhibited mitochondrial fragmentation and NRF2 nuclear accumulation in cells subjected to cytoplasmic irradiation. However, inhibition of mitochondrial fragmentation did not have a significant effect on p53 level.

A-2-3 Mitochondrial superoxide stimulated NRF2 translocation via p53 and mitochondrial fragmentation

Elevation of mitochondrial superoxide was measured using fluorescent probe, MitoSOX in WI-38 cells receiving cytoplasmic irradiation. Scavenging MitoSOX with 10 μM mito-TEMPOL significantly reversed the levels of p53, mitochondrial fragmentation, and NRF2 nuclear accumulation in cytoplasm-irradiated cells. Therefore, a signaling pathway initiated by MitoSOX, with the participation of p53 and its mediating role in mitochondrial morphology, was proposed to contribute to the activation of NRF2 antioxidative response, suggesting its defensive role against cytoplasmic radiation-induced DSB.

A-2-4 Cytoplasmic radiation-induced radioadaptive response

Upon observing the phenomena described above, we went on to investigate whether low dose cytoplasmic irradiation could confer cellular resistance to high dose radiation. Twenty-four hours after cytoplasmic irradiation delivering 500 protons, WI-38 cells were exposed to 2 Gy or 6 Gy broad beam X-ray radiation. Thereafter, the cellular DSB levels were detected. The results showed that the priming cytoplasmic irradiation decreased the DSB levels caused by later exposure to 2 Gy and 6 Gy X-ray irradiation. The phenomenon was negated if cells were pretreated with mito-TEMPOL, cPITO (nitric oxide scavenger), Mdivi-1, or pifithrin-α (p53 inhibitor) before cytoplasmic irradiation, corroborating the above-mentioned results.

The results obtained from the cytoplasmic irradiation research performed in this IOL program indicated the involvement of an NRF2 antioxidative response in alleviating cytoplasmic radiation-induced nuclear DNA damage, and defined the roles of MitoSOX, p53, and changes in mitochondrial morphology in the cellular context. We also observed several interesting phenomena, such as down-regulation of p53 in cytoplasm-irradiated cells. These findings showed that secondary targets in cytoplasm play a significant role in the overall damage incurred by nuclear targets; the elucidation of these secondary targets might provide novel treatment strategies using radiotherapy or radiation protection.

B. Radiation induced bystander effect and inter-cellular communication

B-1 Non-targeted normal bystander cells modulate DNA double strand break repair in microbeam targeted cancer cells through gap junction intercellular communication.

Bi-directional signaling involved in the radiation-induced bystander effect (RIBE) between irradiated carcinoma cells and their surrounding non-irradiated normal cells is relevant to radiation cancer therapy. Using the SPICE-NIRS microbeam, we delivered 500 protons to A549-GFP lung carcinoma cells stably

expressing H2B-GFP, which were co-cultured with normal WI-38 cells. The level of γ-H2AX, a marker for DSB, was subsequently measured up to 24 hours post-irradiation in both targeted and bystander cells. The results showed that inhibition of gap junction intercellular communication (GJIC) attenuated DSB repair in targeted A549-GFP cells and suppressed RIBE in bystander WI-38 cells but not in distant A549-GFP cells. This suggests that GJIC plays a two-way role by propagating DNA damage from carcinoma to normal cells and reversing the bystander signaling, also called the "rescue effect," from bystander cells to irradiated cells, to enhance the DSB repair in targeted cells.

In this study, we have demonstrated the involvement of GJIC in targeted A549-GFP cells and non-targeted WI-38 cells by targeting only the A549-GFP cells in the co-cultured population using microbeam technology. Overall, the results indicate that the GJIC between the cancer cells and normal cells involves not only the propagation of DNA damaging signals, but also contributes to upregulation of the protective response in both irradiated cancer cells and bystander normal cells. These results improve our current understanding of the RIBE and the involvement of GJIC among the two types of cells in radiation cancer therapy.

B-2 The importance of the primary and secondary bystander effects in the cross-talk between human lung cancer and normal lung cells after proton microbeam irradiation

Our main research interests are centered on understanding the role and mechanism of gap junction intercellular communication (GJIC) in determining human response to proton microbeams. Particularly, the focus is to study the secondary bystander signaling events between the primary bystander normal lung cells (WI-38) and proton-irradiated lung cancer (A549) cells. The objective is to gain greater understanding of the mechanism underlying the role of GJIC in the spread of toxic effects in normal tissues after proton-irradiation. The results may provide insight into the potential benefits of cancer radiotherapy with enhanced cancer cell killing effects and substantial protection against the propagation of damaging effects to healthy tissue.

We have shown that proton microbeams induced primary bystander responses in both A549 and WI-38 cells. We also found that the secondary bystander responses are propagated from the primary bystander A549 cells to non-irradiated WI-38 cells using the Transwell® insert co-culture method. Inhibition of GJIC in bystander cells may reduce toxic effects during irradiation. These findings demonstrate that GJIC plays a role in mediating proton-microbeam-irradiation-induced secondary bystander responses, which is directly relevant to cancer radiotherapy. Our ongoing studies with high-LET (linear energy transfer) carbon ions show effects that are similar to those detected with protons.

Activities

1. Exchange history and experiment activities (* supported by IOL)

1) *2015/11/16-2015/12/03 N Autsavapromporn, TAF Tengku Ahmad, discussion and a trial microbeam experiment on the studies of normal-cancer bystander effect. (Photo@SPICE irradiation room)

2) 2015/09/30-2015/10/06 Dr. LF Spitta, Mr. S Diegeler (DLR, Germany), Discussion on the experimental set up for a trial microbeam experiment using their HEK-293 NFkB-GFP cell line. Photo was taken at New Charged Therapy facility (left) and get together dinner (right)

3) 2016/2/01-2016/2/11 Dr. LF Spitta, Prof. CE Hellweg, Discussion on the experimental set up for a trial microbeam experiment using their HEK-293 NFkB-GFP cell line. Also, *2016/01/14-2016/02/13 S Diegeler, First trial experiment with their HEK293 NFkB-GFP cell line. Nucleus and Cytoplasm targeted experiment was performed to see whether their cell line is acceptable for microbeam irradiation.*2015/09/10-2015/10/09: J Wang, SPICE beamtime experiment, supported by IOL

4) 2016/02/18-2016/02/23: Lianyun Chen & Furu Zhan, Discussion about development of microfluid based microbeam facility for cellular radiation research, supported by Chinese Academy of Sciences. Photo at the seminar (left), and in front of SPICE beam line (right)

5) *2016/02/18-2016/02/23: Li-jun Wu, Discussion about microbeam radiation induced biological effects, supported by IOL. A seminar titled “Development of ASIPP microbeam facility and its related biological studies.” By given by Prof. Wu.
6) *2016/02/18-2016/03/08: Jun Wang, SPICE beam time experiment, supported by IOL
7) 2016/5/10-2016/11/8：Lu Dong, Visiting Scholar, Chinese Academy of Sciences Visiting Scholarship.

   Lu Dong joined our SPICE-BIO research core on low dose biological effect and radiation induced bystander effect studies.
8) 2016/11/14 -2016/12/ 21 Dr. N Autsavapromporn (Chiang Mai Univ, Thailand), JSPS fellow
9) *2017/02/27-2017/03/03 J Wang, A Hashim, L Dong, W Punfa, participants of IOL symposium

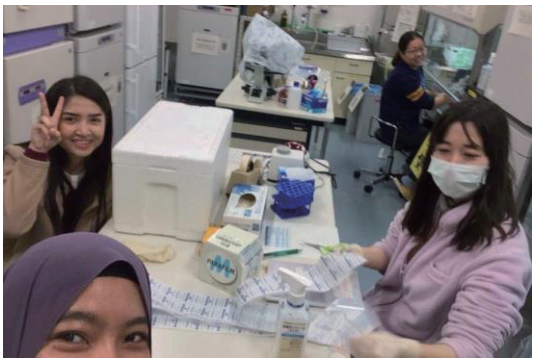
10) 2017/02/10-2017/03/03 N Autsavapromporn (Chiang Mai Univ, Thailand), TAF Ahmad (Malaysia Nuclear Agency, Malaysia) SPICE beam time experiments, IOL symposium
11) T Konishi, Oral presentation@The 5th International Symposium on Space Radiation and Particle Radiotherapy, 2017-05-26; IOL members, T Konishi, J Wang, G Yang, L Dong, L Wu, C Liu, Y Furusawa, and N Autsavapromporn, participated in the symposium.

12) 2017/07/03 – 201707/31 TAF Tengku Ahmad, SPICE beam time experiment.

13) 2017/05/21-2017/05/24 N Autsavapromporn visited Hefei Institute of Physical Science, Chinese Academy, Discussions on the studies of radiation induced anti-oxidative response.


14) 2017/09/11-2018/02/23: N Dukaew (Miw), Studies on radio-sensitization of Eurycomalactone and Its Possible Mechanisms and Radiation Induced Cellular Response Using SPICE Microbeam, MEXT Nuclear Researchers Exchange Program 2017. Miw joined our SPICE-BIO research core. During her stay in NIRS, Professor A Wongnoppavich was invited for discussion on the collaborative studies with Chiang Mai University.

15) 2017/12/11 Professor Ariyaphong Wongnoppavich, Dr. N. Autsavapromporn, Ms. Nahathai Dukaew of Chiang Mai Univ., Dr L Dong of CAS, Dr. C Liu, Ms. A Kobayashi of NIRS, and Group from Fudan University discussed on their progress on the studies related to radio induced bystander effect. Photo is at dinner get-together.

16) 2017/11/05-2017/11/08 T Konishi visited Hefei Institute of Physical Science, Chinese Academy of Science, discussed on microbeam collaborative studies and microbeam facility development. Gave a seminar on Radiobiological Studies with SPICE-NIRS microbeam to Prof. L Wu, J Wang and their member at Hefei Institute of Physical Science. Also had wonderful discussion on the current studies

of Ph. D graduates in Prof. J Wang's lab.

17) * 2017/11/06- 2017/12/20 TAF Tengku Ahmad, SPICE beam time experiment
18) 2017/12/18-2018/05/18: Prof. J Wang, SPICE beam time experiment, supported by China Scholarship Council
19) 2018/01/30 Meeting with Prof Tom Hei, Columbia University @ ANA hotel near NARITA airport. Discussion on our progress and outcomes of SPICE-BIO research core. (Photo, Prof. J Wang, Prof. T Hei, and TK)

20) *2018/05/19- TAF Ahmad, SPICE beam time experiment by TK'S KAKENHI
21) *2018/05/25 – 2018/06/25 N Dukaew, SPICE beam time experiment, supported by IOL
22) *2018/06/14 – 2018/06/19 J Wang, speaker of the IOL final symposium
23) 2018/06/21-2018/07/11 Narongchai Autsavapromporn, SPICE beam time experiment by TK'S KAKENHI

2. Publication and Presentation

2-1) Peer-review publications: (New to Old, from 2015 May)

2-1-1) Submitted

1) Wang J and Konishi T*, NRF2 antioxidative response mitigates cytoplasmic radiation induced DNA double strands breaks., *(submitted to Cancer Science, 2018 May 19th)*
2) Kobayashi A and Konishi T*, Radiation quality affects alteration in COX-2 pathway to trigger radiation induced bystander effect in A549 lung carcinoma cells. *(submitted to Journal of Radiation Research, 2018 April 24th)*
3) Kobayashi A, Autsavapromporn N, Tengku Ahmad TAF, Oikawa M, Homma-Takeda S, Furusawa Y, Wang J, Konishi T*, Bystander WI-38 cells modulate DNA double-strand break repair in

microbeam-targeted A549 cells through gap junction intercellular communication. (2017 Dec. submitted to *Radiation Protection Dosimetry*)

2-1-2) Published

4) Kobayashi A, Tengku Ahmad TAF, Autsavapromporn N, Oikawa M, Homma-Takeda S, Furusawa Y, Wang J, Konishi T* (2017) Enhanced DNA double-strand break repair of microbeam targeted A549 lung carcinoma cells by adjacent WI38 normal lung fibroblast cells via bi-directional signaling. Mutat Res 803-805:1-8. doi:10.1016/j.mrfmmm.2017.06.006
5) Autsavapromporn N*, Liu C, Konishi T (2017) Impact of Co-Culturing with Fractionated Carbon-Ion-Irradiated Cancer Cells on Bystander Normal Cells and Their Progeny. Radiat Res 188 (3):335-341. doi:10.1667/RR14773.1
6) Autsavapromporn N*, Konishi T, Liu C, Plante I, Funayama T, Usami N, Azzam EI, Suzuki M (2017) A correlation of long term effects and radiation quality in the progeny of bystander cells after microbeam radiations: The experimental study of radiotherapy for cancer risk mitigation. Journal of Physics: Conference Series 860:012026. doi:10.1088/1742-6596/860/1/012026
7) Chen N, Zhang R, Konishi T, Wang J* (2017) Upregulation of NRF2 through autophagy/ERK 1/2 ameliorates ionizing radiation induced cell death of human osteosarcoma U-2 OS. Mutat Res 813:10-17 doi:10.1016/j.mrgentox.2016.11.006
8) Oikawa M*, Suya N, Konishi T, Ishikawa T, Hamano T, Homma-Takeda S (2015) Micro-PIXE analysis system at NIRS-electrostatic accelerator facility for various applications. International Journal of PIXE 25 (03n04):217-225. doi:10.1142/S0129083515500187

2-2) Conference presentation

1) Ohsawa D, Furusawa Y, Kobayashi A, Oikawa M, and Konishi T, Analysis of SPICE microbeam profile using fluorescent nuclear track detector (FNTD), ICNMTA　2018, Guildford, Surrey, UK, 2018-07-08-13
2) 小西　輝昭、SPICE-NIRS microbeam が先導する放射線生物学（Single Cell Radio-Biology）量子生命科学研究会第 2 回学術集会 2018 年 5 月 10 日、東京大学弥生行動一条ホール (Oral)
3) Wang J, Konishi T, Kobayashi A, Furusawa Y, Oikawa M, Ohsawa D, Autsavapromporn N, Tengku Ahmad TAFT, Study on Cytoplasmic Radiation Induced Defensive Signaling using SPICE-NIRS microbeam. 量子生命科学研究会第 2 回学術集会 2018 年 5 月 10 日、東京大学弥生行動一条ホール　(poster)
4) Kobayashi A, Tengku Ahmad TAFT, Autsavapromporn N, Oikawa M, Furusawa Y, Konishi T, Analysis of radiation-induced bystander response between human normal cells and cancer cell using SPICE-NIRS microbeam system. 量子生命科学研究会第 2 回学術集会 2018 年 5 月 10 日、東京大学弥生行動一条ホール (poster)
5) 大澤大輔、古澤佳也、小林亜利紗、及川将一、小西輝昭、蛍光飛跡検出器（Al2O3 C,Mg）を用いた SPICE マイクロビームサイズの高精度評価。量子生命科学研究会第 2 回学術集会 2018 年 5 月 10 日、東京大学弥生行動一条ホール (poster)

6) Autsavapromporn N, Kobayashi A, Liu C, Tengku Ahmad TAFT, Dukaew N, and Konishi T, The Secondary Bystander Effect Induced by Proton Microbeam Irradiation. 量子生命科学研究会第２回学術集会 2018 年 5 月 10 日、東京大学弥生行動一条ホール (poster)
7) Kobayashi A, Autsavapromporn N, Tengku Ahmad TAF, Oikawa M, Homma-Takeda S, Furusawa Y, Wang J, Konishi T, Bystander WI-38 normal lung fibroblast cells modulate DNA double-strand break repair in microbeam-targeted A549 cells through gap junction intercellular communication, MICROS 2017 17th International Symposium on Microdosimetry, 2017. 11.7 (Oral)
8) 小西　輝昭, 小林　亜利紗, 大澤　大輔, 劉　翠華, 古澤　佳也, 及川　将一, SPICE-NIRS microbeam：マイクロビームのアドバンテージを活用した放射線生物研究、日本放射線影響学会, 2017-10-27 (oral)
9) 小林　亜利紗, 小西　輝昭, NOS と COX-2 を指標としたバイスタンダー効果因子誘発機序に対する間接作用の寄与の解析. 日本放射線影響学会, 2017-10-27 (ポスター発表)
10) Kobayashi A, Autsavapromporn N, Tengku Ahmad TAF, Homma-Takeda S, Furusawa Y, Wang J, Konishi T, Bi-directional cellular signaling between microbeam targeted cancer cells and non-targeted normal cells, 1st QST International Symposium "Quantum Life Science" -The path breaking life-scientists with quantum eyes and hands. 2017.7.25-26 (Poster)
11) Konishi T, Oikawa M, Kobayashi A, Ohsawa D, Homma-Takeda S, Furusawa Y, Hamano T, SPICE-NIRS microbeam: A focused vertical system for proton irradiation of a single cell for radiobiological research. 1st QST International Symposium "Quantum Life Science" -The path breaking life-scientists with quantum eyes and hands. National Institutes for Quantum and Radiological Science and Technology (QST), 2017-07-25
12) Konishi T, Wang J, Kobayashi A, Autsavapromporn N, Tengku Ahmad TAF, Ohsawa D, Furusawa Y, Studies on defensive cellular response induced　by cytoplasm/nucleus targeted irradiation using SPICE-NIRS microbeam. 1st QST International Symposium "Quantum Life Science" -The path breaking life-scientists with quantum eyes and hands. National Institutes for Quantum and Radiological Science and Technology (QST), 2017-07-25
13) Kobayashi A, Autsavapromporn N, Tengku Ahmad TAF, Oikawa M, Homma-Takeda S, Furusawa Y, Wang J, Konishi T, Study of radiation induced cell response between heterologous cells using proton beam microbeam irradiation method, The 44th Annual Meeting of the Japanese Society of Toxicology, Kanagawa,2017-07-10 (poster)
14) Konishi T, Wang G, Tengku Ahmad TAF, Autsavapromporn N, Kobayashi A, Oikawa M, Furusawa Y, Enhanced DSB repair triggered by cytoplasmic damage induced by proton microbeam irradiation. The 5th International Symposium on Space Radiation and Particle Radiotherapy, 2017-05-26 (Oral)
15) Konishi　T,　Kobayashi A, Furusawa Y, Liu C, Oikawa M, Tengku Ahmad TAF, Autsavapromporn N, Wang J, Microbeam induced cytoplasmic damage triggers activation of DNA double-strand break repair. 日本放射線影響学会, 2016-10-27　（poster）
16) Autsavapromporn N, Konishi T, Liu C, Azzam EI, Plante I, Funayama T, Suzuki M, Late Effects

in the Progeny of Bystander Human Cells after Carbon Ions are Dependent on Radiation Quality: The Relevance to Cancer Risk., Thailand Institute of Nuclear Technology, 2016-08-05 (Oral)

17) Jun Wang, Oral presentation at the 10th Chinese Conference of Radiation and Environmental Biophysics, Quanzhou, China, 2016/04/22-2016/04/26 (Oral)
18) Kobayashi A, Oikawa M, Konishi T, Furusawa Y, Dynamics of radiation induced bystander effect between cancer cells and normal cells, Japan Radiation Research Society 59th Congress, Hiroshima, 2016-10-26 (Oral)
19) Kobayashi A, Konishi T, Oikawa M, Uchihori Y, Takeda S, Kumagai Y, Furusawa Y, An examination of how neighboring un-irradiated normal cells inhibit repair of irradiated cancer cells, The 12th International Workshop on Microbeam Probes of Cellular Radiation Response, Fukui, 2015-06-01 (Poster)
20) Konishi T, Kobayashi A, Yu KN, Yang G, Tengku Ahmad TAF, Oikawa M, Furusawa Y, SPICE-NIRS Microbeam: a focused vertical system for proton irradiation of a single cell for radiation biology. IWM2015, Microbeam Workshop 2015, 2015-05-31 (Oral)
21) Wang J, Konishi T, Kobayashi A, Oikawa M, Hei TK, Uchihori Y, Wu LJ, Activation of Nrf2 Antioxidative Response In Normal Human Lung Fibroblast WI38 By Cytoplasm Targeted Irradiation With Proton Microbeam In NIRS, IWM2015, Microbeam Workshop 2015, 2015-05-31 (Poster)
22) Autsavapromporn N, Plante I, Liu C, Konishi T, Usami N, Funayama T, Azzam EI, Murakami T, Suzuki M, Late Effects in the Progeny of Bystander Human Cells are Dependent on Radiation Quality: The Relevance to Cancer Risk., IWM2015, Microbeam Workshop 2015, 2015-05-31
23) Liu Y, Kobayashi A, Maeda T, Fu Q, Oikawa M, Yang G, Konishi T, Uchihori Y, Hei TK, Wang Y, Target irradiation induced bystander effects between stem-like cells. IWM2015, Microbeam Workshop 2015, 2015-05-31
24) Pandey BN, Desai S, Kobayashi A, Konishi T, Transmission and repair of DNA damage signal to bystander cells from the population of proton microbeam irradiated human cells. IWM2015, Microbeam Workshop 2015, 2015-05-31
25) Konishi T, Kobayashi A, Oikawa M, Furusawa Y, Yu KN, Yang G, Shirakawa Y, Uchihori Y, Current Status and Radiobiological Studies Using SPICE-NIRS Microbeam Irradiation System ICRR2015, Organizing Committee, 2015-05-28
26) Kobayashi A, Konishi T, Oikawa M, Uchihori Y, Takeda S, Kumagai Y, Furusawa Y, Analysis of the bystander effect between microbeam targeted cancer cells and non-targeted normal cells,15th International Congress of Radiation Research, Kyoto International Conference Center, 2015-05-27 (Poster)
27) Autsavapromporn N, Plante I, Liu C, Konishi T, Usami N, Funayama T, Uchihori Y, Hei TK, Azzam EI, Murakami T, Intercellular Communication in the Propagation of Bystander Effect and Genomic Instabilty in Human Cells after X-ray, Proton and Carbon., International Congress of

Radiation Research Committee, 2015-05-28 (poster)

3. Application of external grant fundings

3-1) Teruaki Konishi

1) FY2018(H30) - FY2021, JSPS Grant-in-Aid for Scientific Research A (as one of the 6 participants) [Not accepted]
2) FY-2018 - JSPS, Quantum Life Science Project, World Premier International Research Center Initiative (WPI), T. Konishi (one of the 11 principle investigators for the program) [Not accepted]
3) FY2017(H29) - FY2019, JSPS Grant-in-Aid for Scientific Research B (17H04268) **[Accepted]**
4) FY2017(H29) May., JSPS SAKIGAKE [Not accepted]
5) FY2016(H28) -FY2018, JSPS Grant-in-Aid for Exploratory Research (16K15586) **[Accepted]**
6) FY2015(H28) Nov. JSPS Japan Core to Core program B [Not accepted]
7) FY2013(H26) - FY2015, JSPS Grant-in-Aid for Young Scientists (B) (25861137) **[Accepted]**

3-2) Jun Wang

8) (Submitted) 2019-2023, Key project of Natural Science Foundation Committee of China, Jun Wang (one of the 4 11 principle investigators for the project)
9) (Submitted) 2019-2022, Regular project of Natural Science Foundation Committee of China, Jun Wang (principle investigator)
10) 2017. March: as one of the 4 principle investigators, 2017 March, Key project of Natural Science Foundation Committee of China [Not accepted]
11) 2016. Jun Wang/Teruaki Konishi, (Institutional) International Bi-lateral collaborative program, CAS [Not accepted]
12) 2017-2018, Visiting Scholar to QST-NIRS supported the China Scholarship Council (201704910370) **[Accepted]**
13) 2017-2018, Innovative Program of Development Foundation of Hefei Center for Physical Science and Technology (2016FXCX005) **[Accepted]**
14) (Co-principle investigator): 2017-2019, Key project of International Partnership Program supported by Chinese Academy of Sciences (116134KYSB20160084) **[Accepted]**
15) 2016-2019, Regular project of Natural Science Foundation Committee of China (11575232) **[Accepted]**

3-3) Narongchai Autsavapromporn

16) 2018-2023, National Research Council of Thailand (NRCT)-Chiang Mai University (Submitted). **[Accepted]**
17) 2017-2020, International Atomic Energy Agency (IAEA), (CRP contact number: 21062). **[Accepted]**
18) 2017. The Joint Research Project under JSPS-NRCT. [Not accepted]
19) 2016-2017, National Research Council of Thailand (NRCT)-Chiang Mai University. **[Accepted]**
20) 2016, JSPS Bridge Fellow, (BR161201). **[Accepted]**

21) 2016-2018, The Thailand Research Fund (TRF)- The Office of the Higher Education Commission. **[Accepted]**

3-4) Lijun Wu

22) 2016-2019, Project from Chinese Academy of Sciences for Incorporating microfluid cellular irradiation system into the ASIPP proton microbeam facility **[Accepted]**

3-5) Alisa Kobayashi

23) FY2017(H29) – FY2019, JSPS KAKENHI Grant-in-Aid for Young Scientist B (17K16496) **[Accepted]**

4. Awards, Travel Awards

1) **Alisa Kobayashi**; 2017 Nov 5th – 10th , Financial Assistance for Young Investigators, 17th International Symposium on Microdosimetry, Venice (Venezia), Italy, Congress Center - "Cultural Center Don Orione Artigianelli", Zattere -Dorsoduro 919 - 909/A, 2017.11.5- 10.

5. Beam time proposals for collaborative research (SPICE/HIMAC and other facilities)

SPICE-NIRS microbeam proposals

1) FY2016-FY2018, PI: Teruaki Konishi, SPICE proposal # S16-IOL01, "Studies on Radiation Induced defensive cellular signaling using SPICE-NIRS microbeam."
2) FY2016-FY2018 PI: Narongchai Autsavapromporn, SPICE proposal # S16IOL02, "The Importance of the Primary and Secondary Bystander Effects Cross-Talk between Human Lung Cancer and Lung Normal Cells after Proton Microbeam Irradiation"
3) FY2016- FY2017, PI: Alisa Kobayashi, SPICE proposal #S16-AK01, "Analysis of bystander response between cancer cells and normal cells"
4) FY2018- FY2020 PI: Daisuke Ohsawa, SPICE proposal # S18-DOH01, "Studies on the kinetics of DSB repair proteins by spatiotemporal regulation of DNA damage complexity using SPICE microbeam"

HIMAC-NIRS proposals

5) FY2016- FY2017, PI: Narongchai Autsavapromporn, HIMAC proposal # 16J318, "The Importance of Primary and Secondary Bystander Response Crosstalk between Human Lung Cancer and Normal Cells by Exposure of High-LET Carbon Ions".
6) FY2017- FY2019, PI: Alisa Kobayashi, HIMAC proposal # 17J328, "Analysis of dose and LET dependence of carbon ion induced bystander response by COX-2 induction"

6. Others, MOU/Cooperative agreements.

1) 2018 March, MOU- between Columbia University and QST
2) 2017 October, MOU- between Faculty of Medicine, Chiang Mai University and National Institute of Radiological Sciences

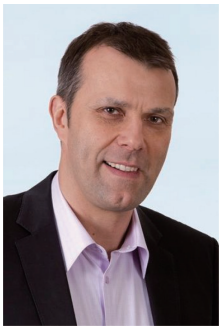
Radiation damage mechanisms at molecular level approached with physicochemical technologies

CORE LEADER (J)

Satoshi Kodaira kodaira.satoshi@qst.go.jp
National Institute of Radiological Sciences (NIRS) / QST

Satoshi Kodaira is a Principal Research Scientist of the Radiation Measurement Research Team at NIRS. He received his Ph.D. degree from Waseda University, Japan in 2007. His current research is on radiation dosimetry in mixed radiation. He has extensive experience in the heavy ion beam research field and has contributed many publications and reviews. He received the Zeldovich Medals from COSPAR on August 2016. He dedicates to the social activity as an associate editor of the Life Sciences in Space Research and the International Committee of the International Nuclear Track Society.

CORE LEADER (F)



Rémi Barillon remi.barillon@iphc.cnrs.fr
Institut Pluridisciplinaire Hubert Curien (IPHC)

Rémi Barillon is Professor at the Faculty of Chemistry of the University of Strasbourg (France) and Director of IPHC/CNRS. He studies interaction between ionizing rays and organic matter and its applications to dosimetry (radioprotection, nuclear physic and radiotherapy). He was co-chairman of the 27th International Conference on Nuclear Tracks and Radiation Measurements (Strasbourg, 2017) and he is the Secretary of the International Nuclear Track Society.

Core Members

- Quentin Raffy, Ph.D., Jean Marc Jung, Ph.D., Nicolas Ludwig, Ph.D. student, Philippe Peaupardin, Ph.D., Catherine Gaindo, Ph.D., Christelle Roy, Ph.D., Marc Rousseau, Ph.D., Ziad El Bitar, Ph.D., Abdel Mjid Nourreddine, Ph.D., Nicolas Arbor, Ph.D. (IPHC/CNRS)
- Michel Fromm, Ph.D, Jean-Emmanuel Groetz, Ph.D (Universite de Bourgogne Franche-Comte)
- Gerard Baldacchino, Ph.D., Behnaz Behmand, Ph.D., Cecile Sicard-Roselli, Ph.D. (CEA/CNRS)
- Teruaki Konishi, Ph.D., Tamon Kusumoto, Ph.D., Mitsumasa Taguchi, Ph.D., Shinji Sato, Msc. Masakazu Oikawa, Msc. (QST)
- Tomoya Yamauchi, Ph.D. (Kobe Univ.)

Purpose of the Project:

This project aims to understand the radiation damage mechanism at molecular level, especially of organic matter or related material, caused by heavy ions. We survey the whole stages from the initial process due to the ionization, the chemical reactions associated with OH radicals, and permanent damage along the trajectory in polymers in solid form and in amino acids in aqueous solutions or proteins. The inter-comparison of the

physical quantities related to LET (linear energy transfer), absorbed dose, the chemical yields and cross sections of hydroxyl radicals (in case of aqueous solutions) and permanent damage in the polymers and biomolecules will give new insights to describe radiation effect at the molecular level. Thus, obtained experimental data will contribute to the improvement of the Geant4-DNA Monte-Carlo simulation as benchmark.

Summary of the Project:

We planned to employ two kinds of approaches of radiation physics and chemistry technologies. The first one is visualizing fluorescent signals along a heavy ion trajectory by a confocal microscopy. We have already observed fluorescent signals of both an ion track and a damaged location in cells cultured on the Al_2O_3:C,Mg crystal. We tried to observe a fluorescent track in gel containing fluorescent probe reacting with hydroxyl radicals for co-visualizing an ion track and a scavenged location. Unfortunately, low fluorescence intensity prevented us from successful observation in gel. The second approach is the radiolysis study measuring radiation induced radicals in aqueous solution and permanent damages in solids. This approach was successfully carried out for collecting the fundamental data in 1) biomolecules (amino-acid (phenyl alanine) and proteins in aqueous solution) and 2) polymer in a solid form as follows.

1) Biomolecules

Since water is a major component of biological cells (~65%), and, therefore, absorb most of the energy deposited by ionizing radiations, water radiolysis is indispensable to understand the radiation effect on a living organism. Among the products formed by radiation, hydroxyl radical $HO^{\bullet}$ is one of the most potent species causing molecular damages. In particular, proteins, of which amino-acids are the elementary building blocks, represent about 20 % of the cell in mass. Yet, the literature on their fate under ion irradiation is very scarce.

Our first studies focused on an aromatic amino-acid and a small protein. For the amino-acid, we chose phenylalanine, which had been thoroughly examined under gamma-ray irradiation. It is also one of the very few amino acid studied under heavy-ion irradiation to this date, in one publication. As for the protein, myoglobin was chosen, a small heme-protein which structure consists mainly of alpha helices. Apo-myoglobin, that is myoglobin without its heme, was also considered, to assess the influence of the heme moiety.

1-1) Water radiolysis

As the first trial in 2016, we performed the determination of the kinetics of $HO^{\bullet}$ yields with protons of various energies, between 2.5 and 5 MeV at HIMAC, which was compared with the results obtained by IPHC using other facility in Strasbourg, a 4 MV electrostatic accelerator. A scavenger molecule was employed to measure the amounts of hydroxyl radical formed under irradiation of aqueous solutions. 3-carboxylic coumarin acid (3CCA) is known to react very quickly and selectively with $HO^{\bullet}$ ($k = 6.9 \times 10^9$ $M^{-1}.s^{-1}$) to yield multiple products, among which 7-Hydroxy-3-carboxylic coumarin acid (7OH-3CCA) is the most fluorescent

one. Since the yield of formation of 7OH-3CCA is precisely known, determination of the quantities of 7OH-3CCA formed by fluorescence measurements gives access to the amount of $HO^{\bullet}$ generated by water radiolysis. The characteristic scavenging time depending on the 3CCA concentration allows the reconstruction of $HO^{\bullet}$ yields kinetics, from 7 ns to ~1500 ns after passage of the incident particle.

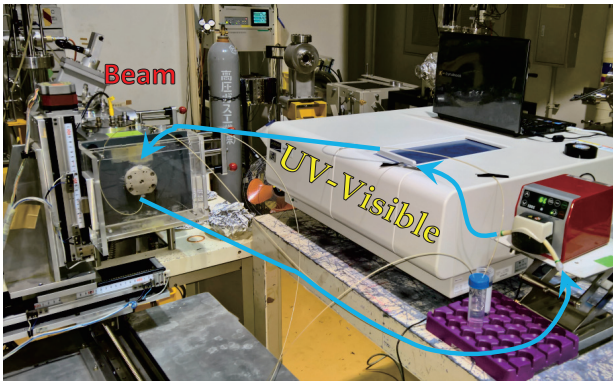


Figure 1: Irradiation set-up with flow-through irradiation cell circulating loop at HIMAC-MEXP.

The solutions were circulated in a closed loop, and irradiated in a flow-through cell specifically designed as shown in **Figure 1**. The circulation allows homogeneous irradiation, and UV-Visible and/or fluorescence online measurements. The irradiated solutions were analyzed by HPLC after irradiation, using an inverse phase column and a Hitachi fluorimeter as fluorescent detector. Calibration curves prepared with commercial 7OH-3CCA allowed determination of the coumarin concentrations.

At the beginning of IOL program, this core possessed no analysis equipments such as fluorescence spectrometer, UV/Visible spectrophotometer, HPLC and so on. Since the IOL program covers only travel expense for meeting and experiment, there was no chance to purchase these equipments. After intense search, we found an old HPLC which was left unused for long years (maybe about 15 years) in the electrostatic accelerator facility at NIRS. We reconstructed, repaired, and adjusted it, and finally we succeeded to operate it!

The data consistency was found very good, which validates the irradiation setup, as well as the analysis system developed in Chiba using an HPLC and a fluorimeter as detector. The number of hydroxyl radical formed per proton was calculated from the experimental data obtained after HPLC analysis of the irradiated solutions as shown in **Figure 2**. The good consistency between the data obtained at HIMAC and IPHC 4 MV electrostatic accelerator was successfully found. As shown in **Figure 3**, the differential yields G' of $HO^{\bullet}$ were

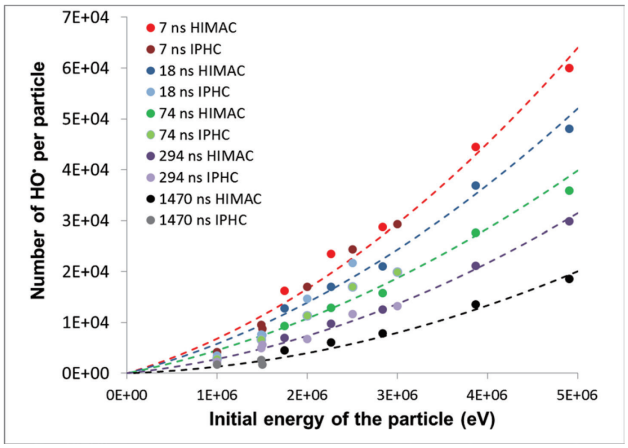


Figure 2: Number of hydroxyl radical formed for each particle. Solid dots: experimental data obtained at HIMAC and at IPHC. Dashed lines: Quadratic fits of the data.

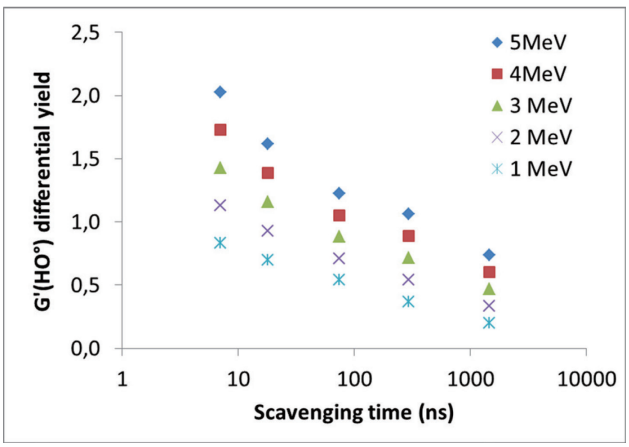


Figure 3: Reconstructed kinetics of the $HO^{\bullet}$ differential yields estimated for proton energies between 1 and 5 MeV.

estimated, that is the yields for infinitesimal energy losses of the particles in water. The yields decrease with time can be explained by the recombination of radical species that occurs in the ion track. In the ns-µs time scale, during the non homogeneous chemistry stage, hydroxyl radical quickly reacts with identical and other radicals to yield molecular species. For each scavenging time, the yield increases with the energy of the proton, which is closely related with the density of the energy deposited by the particle, measured by LET. The highest the energy of the proton, the lowest the LET, and therefore the lowest density of radicals in the ion track, leading to less recombination of $HO^{\bullet}$.

1-2) Amino-acids and proteins

Using the irradiation and analysis systems as we confirmed in 2016FY, the phenylalanine diluted solution (2 mM) was irradiated to protons, helium and carbon ions in aerobic conditions, and the resulting radiolysis products were analyzed by HPLC at HIMAC, and LC-MS at IPHC. Calibration curves had been made at IPHC, and were used for quantitative analyses of the products. Myoglobin and apo-myoglobin diluted solutions (50 µM) were irradiated with carbon and helium ions, under the same conditions. The resulting products were analyzed by direct LC-MS, LC-MS after trypsic digestion, SDS-PAGE and circular dichroism. All analyses were performed at IPHC. Myoglobin concentrated gels (21 %) were cast as 5 µm thick films, sandwiched between two polyethylene 5 µm films. A specific irradiation cell has been developed by IPHC to allow on line infrared spectroscopy (with portable Bruker spectrometer) under irradiation, and in controlled atmosphere. This allowed to observe continuously the structure changes of the proteins during irradiation. Concentrated gels mimic the protein concentration in the cell and show both direct and indirect effects (through water radiolysis products) of the ions.

As already described under gamma rays irradiations, ortho-, meta- and para- tyrosines are also formed under ion irradiation of phenylalanine solutions, by hydroxylation of the amino-acid in oxygenated medium. Radiolysis with various energies allowed determination of the differential yields of these tyrosines, presented on **Figure 4**. With higher LET, the hydroxyl radical density increases also, leading to more radical-radical recombination. This explains the decrease of tyrosines yields with LET. Thus obtained data are quite consistent with literature as well as our data with gamma-rays. Other than tyrosines, several radiolysis products were identified in significant quantities, and in particular di-hydroxy phenylalanines and dimers are worth to be noted. These products had never been reported, neither under gamma rays irradiation nor in the study of Taguchi et al. They were indeed found to be formed in very small quantities with experiments performed under gamma-rays. Therefore, they seem to be species specific to ion radiolysis, forming because of local high radical density.

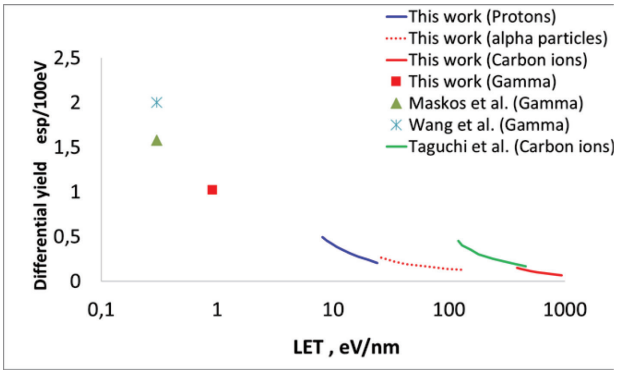


Figure 4: Differential yields of the tyrosines under irradiation of diluted phenylalanine solutions, as a function of LET of particles.

Circular dichroism analyses of myoglobin solutions irradiated by carbon ions showed that the protein loses its alpha helices structure with increasing dose. Comparisons with the apo-myoglobin show that the heme plays a central role in the radiolysis damages, as these are observed for much lower doses with apo-myoglobin compared with myoglobin. Hydroxylated digestion peptides were identified, containing methionine and tryptophan, amino-acids known to be especially sensible to hydroxylation. Measurements of infrared spectra of myoglobin gels under irradiation allowed precise determination of the protein structure changes. The observed behavior is very different from that in diluted solutions, with formation of a specie of a new defined structure. In February 2018, experiments were also performed with high-energy protons and carbon-ions, for phenylalanine, myoglaobine, apo-myoglobine and new target proteins. The analysis of the products is in progress.

2) Polymer

In addition to the water radiolysis, we investigated the mechanism of forming latent track (damaged trail) induced by energetic ions in poly(ally diglycol carbonate) (PADC), which is employed as a solid state nuclear track detector (SSNTD). We performed systematic measurement of chemical cross sections modification under ion irradiation. As secondary electrons created by the incoming ions are responsible for the scission of bonds, ion irradiations have been completed with direct electrons irradiation. **Figure 5** represents the relative absorbance of typical functional groups in PADC irradiated with 28 MeV electrons (left axis). The relative absorbance of ether decreases linearly with increasing electron fluence. Conversely, that for carbonate ester (C=O and C-O-C) and CH groups begin to decrease above the critical fluence of 1.6×10^{15} electrons/cm^2 where is equivalent to the critical dose of 50 kGy. Around the critical fluence, the generation rate of OH groups decreases suddenly. Similar trends are observed with gamma rays, X-rays and UV photons with a wavelength of 222 nm. These findings, named Multi-hit model, imply that more than two electrons are necessary to cleave the carbonate ester in PADC. Namely, the number of secondary electrons is strongly related to the creation mechanism of latent tracks in PADC. Therefore, we calculated the spatial distribution of the number of secondary electrons created by incoming ions using Geant4-DNA to express the detection threshold for etch pit formation in PADC. In the present case, we count the electrons passing through cylinder surface with a certain radius because the tracks of secondary electrons are cylindrically symmetric along the ion path. We set-up a series of cylinders, with radius increasing in steps of 0.1nm, and evaluated the number of electrons that crossed the sides of each cylinder in the both directions, forward and backward. We propose a new index of Radial Electron Fluence

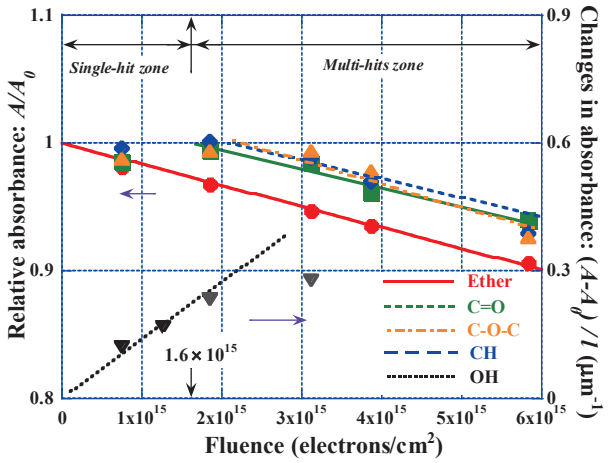


Figure 5: The relative absorbance of typical functional groups in PADC irradiated with 28 MeV electrons.

around Ion Tracks (REFIT) for scaling the detector response as SSNTD instead of conventional dE/dx (i.e. LET). As shown in **Figure 6**, REFIT is defined as the number of secondary electrons which pass through the cylinder surface coaxial to the ion trajectory for proton, He and C ions against the distance from ion's path. The values of REFIT are calculated at each detection threshold. The values of REFIT at a radius of 1 nm, which is the half of the length of a repeat unit of PADC, are in agreement with the Multi-hit model. **Figure 7** shows the track response data as a function of the REFIT at a radius of 1 nm (a). The detection thresholds for each ion are indicated by the arrows in **Fig. 7**. Compared to the response curve in **Fig. 7(b)**, that is expressed as a function of the stopping power, those in **Fig. 7(a)** are closer each other. From this, we judge that the REFIT is an improved physical parameter relative to the stopping power. To improve the accuracy of the simulation of the REFIT, we should address following three major issues:

- In the present simulation, all secondary electrons are ejected from the ion trajectory. However, the primary ionization could occur at atoms distant from the trajectory. In the case of C ions, even electrons at 5.4 nm from the track center are affected. This implies that when impact parameter is considered in the simulation, the value of REFIT around track center will be lower than in the present case.
- The electrons with energies less than 7.4 eV, which is the cut-off energy in the simulations, can also act to cleave the chemical bonds by processes such as dissociative electron attachment. We should thus consider the interactions of such low energy electrons in order to more accurately determine secondary electron histories and the density of such electrons around the ion's trajectory.
- A more physically realistic model for PADC should be used as the target instead of the virtual stopping media. In the materials that contain carbon atoms is a mandatory next step to improve the REFIT.

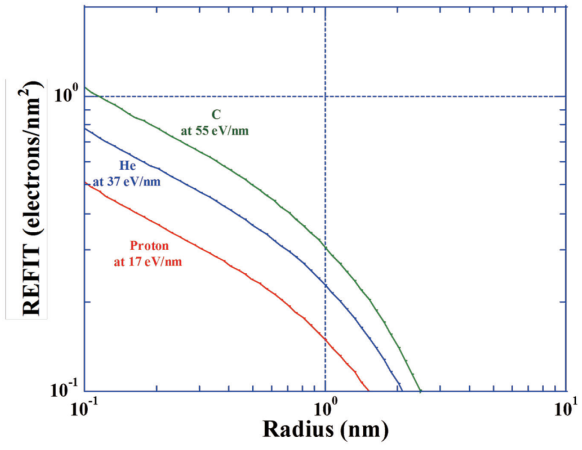


Figure 6: Radial Electron Fluence around Ion Tracks against the distance from ion's path calculated using GEANT4-DNA.

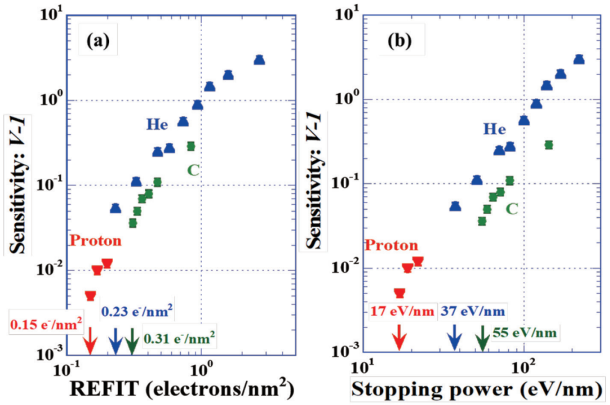


Figure 7: Track response data as a function of the REFIT at a radius of 1 nm (a) and the stopping power (b). The arrows indicate the detection thresholds for PADC for each ion.

Activities:

1) Exchange history and experiment activity

- Kick-off meeting at France [2015, Sep. 13-20th]

- Japan-side (Kodaira, Yamauchi and Kusumoto, Ph.D student) visited IPHC (Strasbourg) and UBFC (Besançon) for discussing about planning of IOL project.
- We had experiment with 1.5 keV soft X-ray irradiation to CR-39 film at UBFC

- Collaboration meeting at Chiba [2016, Mar. 5-10th]
  - France-side (Barillon, Raffy, Roy and Rousseau) visited NIRS (Chiba) for discussing about details of experiment
- IOL joint seminar "*Radiation Damage Detection in Polymers/Biomolecules and Imaging in Particle Therapy* " at Chiba [2016, Mar. 8th]
- HIMAC experiment (H 6 MeV) [2016, Oct. 24-31th]
  - France-side (Barillon, Raffy Ludwig) visited NIRS for the 1st run experiment and discussion.
- HIMAC experiment (H 6 MeV, C 6 MeV/n) [2017, Feb. 19-26th]
  - France-side (Barillon, Raffy, Ludwig) visited NIRS for the 2nd run experiment and discussion
- Meeting at Chiba [2017, Feb. 27th]
  - Discussion of research progress and planning the next experiments (**Figure 8**)

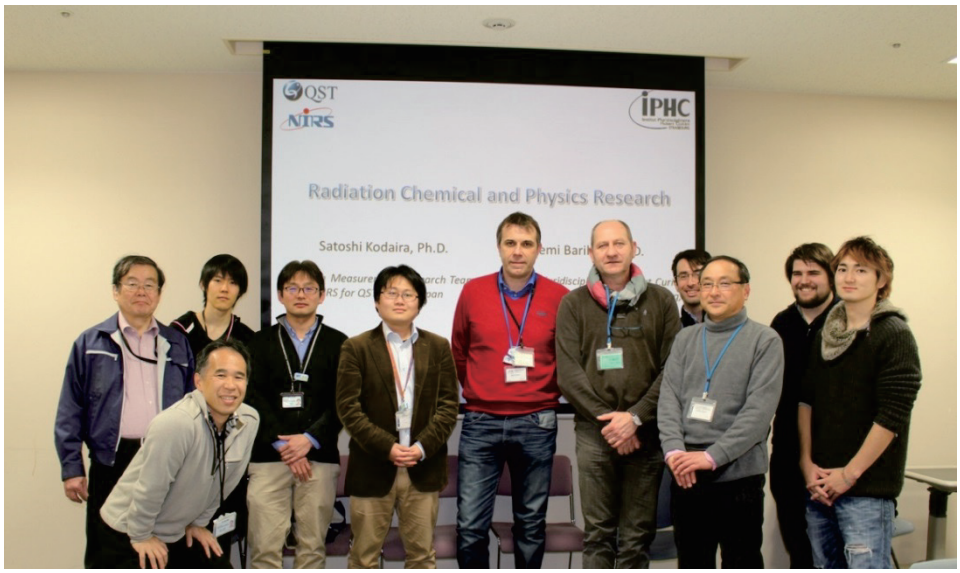


Figure 8: Group photo at the closed meeting on 2017 Feb. 26th at NIRS (Chiba)

- HIMAC experiment (C 6 MeV x2) [2017, Jun. 26th – Jul. 10th]
  - France-side (Raffy, Ludwig, Peaupardin) visited NIRS for the 3rd run experiment and discussion
- Meeting at France [2017, Aug. 26th – Sep. 2nd]
  - Japan-side (Kodaira, Yamauchi, Kusumoto) visited Univ. Strasbourg for the International Conference and had discussion the progress of data analysis of HIMAC experiments
- PhD defense at Kobe Univ. [2017, Dec. 15th]
  - The member, Kusumoto, PhD student at Kobe Univ. and Univ. Strasbourg defended the PhD degree.
  - France-side (Barillon and Fromm) visited Kobe Univ. for the PhD defense and discussion about the future program of joint external grant application
- HIMAC experiment (He 6 MeV/ x3, H 230 MeV, C 400 MeV/n) [2018, Feb. 8 – 23th]

- France-side (Raffy, Batchahane, Baldacchino, Sicard-Roselli, Behmand) visited NIRS for the 4th run experiment and discussion
- Ludwig stayed 2 months from Jan. 9th to Mar. 8th as JSPS research fellow

- HIMAC experiment (He 6 MeV/, C 6 MeV/n x2) [2018, Jun. 8– 23th]
  France-side (Barillon, Raffy, Galindo) visit NIRS for the 5th run experiment and discussion

2) Publication and presentation

2-1) Publication

[1] N. Ludwig, T. Kusumoto, C. Galindo, P. Peaupardin, D. Muller, T. Yamauchi, S. Kodaira, R. Barillon, Q. Raffy, "Radiolysis of Phenylalanine in solution with Bragg-Peak energy protons", Radiation Measurements, 116 (2018) 55-59..

[2] T. Kusumoto, Z. EL Bitar, S. Okada, P. Gillet, N. Arbor, M. Kanasaki, Y. Mori, K. Oda, A-M. Nourreddine, H. Kurashige, M. Fromm, P. Cloutier, A. D Bass, L. Sanche, R. Barillon, T. Yamauchi, "Radial electron fluence around ion tracks as a new physical parameter for the detection threshold of PADC using Geant4-DNA", Radiation Measurements, 118 (2018) 50-53.

[3] T. Kusumoto, M. Fromm, P. Cloutier, A.D. Bass, L. Sanche, R. Barillon, T. Yamauchi, "Elucidation of the Two-Step Damage Formation Process of Latent Tracks in Poly(allyl diglycol carbonate), PADC: Role of Secondary Low-Energy Electrons", The Journal of Physics and Chemistry C (2018) accepted. DOI: 10.1021/acs.jpcc.8b05341.

[4] T. Yamauchi, T. Kusumoto, T. Ueno, Y. Mori, M. Kanasaki, K. Oda, S. Kodaira, R. Barillon, "Distinct step-like changes in G values for the losses of typical functional groups in poly(ethylene terephthalate) along boron ion tracks around the detection threshold", Radiation Measurements, 116 (2018) 51-54.

[5] M. Fromm, S. Kodaira, T. Kasumoto, R. Barillon, T. Yamauchi, "Early processes of nuclear track formation in PADC: the role of intermediate species in the dynamics and formation of a latent track", Materials Chemistry and Physics, (under review).

[6] 楠本多聞, 森豊, 金崎真聡, 小田啓二, 山内知也, 誉田義英, 藤乗幸子, ミッシェル・フロム, ジョン-エマニュエル・グロエ, 小平聡, 北村尚, レミ・バリオン, 「PADC 飛跡検出器の放射線高感受性部に見られる段階的な損傷形成」, 放射線化学, 103 (2017) 41-45.

[7] 川嶋元, 小平聡, 井原大輔, 安田仲宏, 楠本多聞, 森豊, 山内知也, 小林啓一, Eric Benton,「分子長の異なるモノマーから合成した固体飛跡検出器の開発と重粒子線による損傷解析」, 放射線, Vol. 42, No. 3 (2017) 83-90.

[8] 楠本多聞, 寺下佳孝, 森豊, 金崎真聡, 小田啓二, 山内知也, 小平聡, 北村尚, 誉田義英, 藤乗幸子, ジアッド・EL・バイタ, ニコラ・アーバ, クアンタン・ラフィ, レミ・バリオン, ジョン-エマニュエル・グロエ, ミッシェル・フロム, 「高感度飛跡検出器 PADC 中に形成されるイオントラックの特徴」, 放射線, Vol. 42, No. 3 (2017) 73-82.

[9] T. Kusumoto, Y. Mori, M. Kanasaki, K. Oda, S. Kodaira, Y. Honda, S. Tojo, R. Barillon, T. Yamauchi, "Sudden Increase of the Radiation Chemical Yield for Loss of Carbonate Ester in PADC Detector where

the Track Overlapping of 28 MeV Electrons Becomes Significant", JPS Conference Proceedings, 11 (2016) 010001-1-6, doi: 10.7566/JPSCP.11.010001.

[10] T. Kusumoto, Y. Mori, M. Kanasaki, R. Ikenaga, K. Oda, S. Kodaira, H. Kitamura, R. Barillon, T. Yamauchi, "Radiation chemical yields for the losses of typical functional groups in PADC films for high energy protons registered as unetchable tracks", Radiat. Meas., 87 (2016) 35-42.

2-2) Presentation

[1] N. Ludwig, T. Kusumoto, C. Galindo, P. Peaupardin, Y. Le Gall, D. Muller, S. Kodaira, R. Barillon, Q. Raffy, Study of the radiolysis of aromatic amino acids under ion irradiation. 27th International Conference on Nuclear Tracks and Radiation Measurements (ICNTRM), 2017, Strasbourg, France.

[2] T. Kusumoto, Z. EL Bitar, S. Okada, P. Gillet, N. Arbor, M. Kanasaki, Y. Mori, K. Oda, A-M. Nourreddine, H. Kurashige, M. Fromm, P. Cloutier, A. D Bass, L. Sanche; Radial electron fluence around ion tracks as a new physical parameter for the detection threshold of PADC using Geant4-DNA toolkit; 27th International Conference on Nuclear Tracks and Radiation Measurements (ICNTRM), 2017, Strasbourg, France

[3] T. Yamauchi, T. Kusumoto, Y. Mori, M. Kanasaki, K. Oda, S. Kodaira, R. Barillon; Distinct step-like changes in G values for the losses of typical functional groups in poly(ethylene terephthalate) along boron ion tracks around the detection threshold; 27th International Conference on Nuclear Tracks and Radiation Measurements (ICNTRM), 2017, Strasbourg, France

[4] T. Kusumoto, K. Kuraoka, Y. Mori, M. Kanasaki, K. Oda, S. Kodaira, Y. Honda, S. Tojo, R. Barillon, T. Yamauchi; Anomalous increase of the contact angle of water droplets on the surface of PADC detector exposed to proton; 27th International Conference on Nuclear Tracks and Radiation Measurements (ICNTRM), 2017, Strasbourg, France, Poster

[5] T. Yamauchi, T. Kusumoto, K. Azuma, T. Otani, M. Sakai, Y. Mori, M. Kanasaki, K. Oda, S. Kodaira, R. Barillon; Dependence of G values for losses of typical functional groups along heavy ion tracks in bisphenol A polycarbonate on the surface density; 27th International Conference on Nuclear Tracks and Radiation Measurements (ICNTRM), 2017, Strasbourg, France, Poster

[6] N. Ludwig, T. Kusumoto, C. Galindo, P. Peaupardin, Y. Le Gall, D. Muller, T. Yamauchi, S. Kodaira, R. Barillon, Q. Raffy; Determination of radiochemical yields of hydroxyl radical production in water under low energy proton irradiations; 2nd International Symposium on Radiation Detectors and Their Uses (ISRD 2018), 2018, KEK Tsukuba Campus, Japan.

[7] T. Kusumoto, A. Yoshida, K. Yoshida, T. Kambara, Y. Yanagisawa, S. Kodaira, K. Oda, R. Barillon, T. Yamauchi, Applicability of polyimide films for identification of ultra-heavy components, including uranium ions in galactic cosmic ray; 2nd International Symposium on Radiation Detectors and Their Uses (ISRD 2018), 2018, KEK Tsukuba Campus, Japan.

[8] N. Ludwig, T. Kusumoto, C. Galindo, P. Peaupardin, Y. Le Gall, D. Muller, T. Yamauchi, S. Kodaira, R. Barillon, Q. Raffy; Molecular study of the radiolysis of a model protein irradiated by accelerated ions; 30th Miller Conference on Radiation Chemistry, 2017, Castellamarre del Golfo, Italy, Poster.

[9] N. Ludwig, T. Kusumoto, C. Galindo, P. Peaupardin, Y. Le Gall, D. Muller, T. Yamauchi, S. Kodaira, R. Barillon, Q. Raffy; Phenylalanine radiolysis under ion irradiation: Mechanistic study and determination of the radiolytic yields; 30th Miller Conference on Radiation Chemistry, 2017, Castellamarre del Golfo, Italy, Poster.

[10] T. Kusumoto, Y. Mori, M. Kanasaki, K. Oda, Y. Honda, S. Tojo, S. Kodaira, H. Kitamura, R. Barillon, T. Yamauchi; Radiation chemical yields for loss of carbonyl bonds in poly(allyl diglycol carbonate) and other polymeric etched track detector at the LETs ranging from 0.025 to 12,000 keV/μm, International Symposium on Radiation Detectors and Their Uses (ISRD 2016), 2016, KEK Tsukuba Campus, Japan.

2-3) Thesis

[1] Ludwig Nicolas, PhD thesis, "Study of the effect of accelerated ions on amino-acids and proteins", to be defended September 2018, University of Strasbourg.

[2] Kusumoto Tamon, PhD thesis, "Radial Electron Fluence around Ion Tracks as a New Physical Concept for the Detection Threshold of PADC Detector ", Dec. 15th, 2017, Kobe University and University of Strasbourg.

[11] 寺下佳孝, 「メチン基とメチレン基に着目した PADC 中潜在飛跡構造の研究」, 修士論文, 神戸大学大学院海事科学研究科, 2018 年 3 月.

[12] 井原大輔, 「Poly(allyl diglycol carbonate) に生じる放射線損傷の検出法の研究」, 修士論文, 東邦大学大学院理学研究科, 2017 年 3 月.

[13] 上田隆裕, 「赤外顕微鏡を用いた高分子イオントラックの分析」, 修士論文, 神戸大学大学院海事科学研究科, 2017 年 3 月.

[14] 上野琢也, 「重イオン弁別型飛跡検出器開発のための PET の特性評価」, 修士論文, 神戸大学大学院海事科学研究科, 2017 年 3 月.

[15] 亀田結貴, 「PADC 検出器中に存在するヒドロキシル基の定量分析とその化学エッチング特性」, 修士論文, 神戸大学大学院海事科学研究科, 2017 年 3 月.

[16] 池永龍之介, 「顕微赤外分光法によるポリイミド薄膜に対する重イオン照射効果」, 修士論文, 神戸大学大学院海事科学研究科, 2016 年 3 月.

[17] 安田修一郎, 「ポリイミド系エッチング型飛跡検出器の重イオンに対する検出閾値」, 修士論文, 神戸大学大学院海事科学研究科, 2016 年 3 月.

3) Application of external grant funding

[1] Ludwig, JSPS Foreign Research Fellowship (short-term), 2018 Jan. 9th – Mar. 8th, 140,000yen [**Accepted**]

[2] Kodaira, JSPS KAKENHI Young Research (A) 22 million yen, 2017FY-2020FY [**Accepted**]

[3] Kodaira, JSPS KAKENHI Challenging Exploratory Research, 2017FY [Not accepted]

[4] Kodaira, JSPS KAKENHI Young Research (A) [Not accepted]

[5] Raffy, French Program "ANR Jeune Chercheuse – Jeune chercheur" (National research agency, young researcher), 2016FY [Not accepted]

[6] Raffy, French Program “ANR Jeune Chercheuse – Jeune chercheur” (National research agency, young researcher), 2017FY [Not accepted]

[7] Yamauchi, JSPS KAKENHI Scientific Research (C) [16K05002] 3.8 million yen 2016-2018FY [ACCEPTED]

[8] Barillon, French Program “Plan Cancer 2016” 250k€ 2016-2018 [**Accepted**]

[9] Kodaira, JSPS KAKENHI Challenging Exploratory Research, 3.9 million yen, 2015-2016FY [**Accepted**]

[10] Yamauchi, JSPS KAKENHI Scientific Research (C) 2015FY [Not accepted]

[11] Yamauchi, International Academic Exchange Program, Kobe Univ. , 2016-2019 1.4 million yen [**Accepted**]

[12] Yamauchi, Academic Exchange Program for strengthening of relation among Kobe Univ. and QST, 2016, 197,800 yen [**Accepted**]

Impact of heavy ion irradiation on stem cells and tissue regeneration

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Core Members

Ichio Aoki (QST/NIRS)
Viljar Jaks (University of Tartu, Estonia)
Xiaoyan Sun (Karolinska Institutet, Sweden)
Walter Tinganelli (Trento Institute for Fundamentals Physics Applications (TIFPA), Italy)
Alexander Helm (TIFPA, Italy)
Antonella Motta (Centre for integrative biology (CIBIO), Italy)
Dr. Walter Bonani (CIBIO, Italy)

Purpose of the Project:

There are many mechanisms and factors impacting cancer initiation and cancer growth promotion. In 1986 for example, Dvorak *et al* have suggested that tumors are wounds that do not heal. Recently Meng et al supported this theory with the proposition that wound healing is likely one of the major cancer mechanism (Fig 1). Thus, cancer may also be seen as a “natural process related to

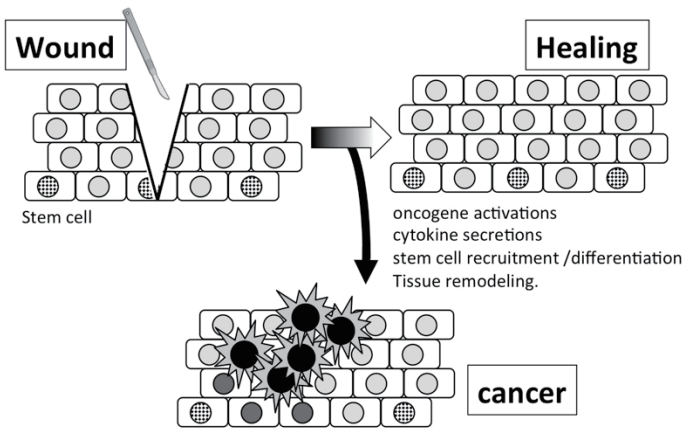


Fig 1. Wound healing as the major cancer mechanism

wound healing", which includes oncogene activations, cytokine secretions, stem cell recruitment differentiation, and tissue remodeling.

In the Heavy Ion Medical Accelerator in Chiba (HIMAC) at the National Institute of Radiological Sciences (NIRS), biological effects of C-ion irradiation using culture cells and animal models have been widely investigated. Whole body irradiation experiments have been performed to study radiation-induced carcinogenesis, and irradiated time-dependent risk of developing cancer was reported. On the other hand, local irradiation also utilized to investigate adverse reactions at short- and long-term, but *de novo* tumor formation was not investigated.

If wound healing is a major determiner for primary and also secondary cancers after radiation treatment, there are no satisfactory models coming from previous reports to evaluate cancer-causing risks, and to investigate the underlying mechanisms of *de novo* cancer up-come due to clinical exposure. To clarify the process after irradiation, it is important to establish a new experimental model, which focuses on the dynamic changes of normal tissues juxtapose to the irradiated area. Specifically, the recruitment of stem cells with increased mutational load due to irradiation might have a crucial role for second-cancer development. Moreover, investigation of the radiation induced wound repair process, compared with mechanical injury, may support our understanding of homeostatic stem cell function in multicellular organisms.

Summary of the Project

This core's activity, in consultation with Dr. Maria Kasper and Dr. Viljar Jaks of the Karolinska Intitutet group (Sweden and Estonia), was supposed to begin in September, 2015. Unfortunately, just after the start of the project, it is found that the time course of the research plan must be drastically altered due to mainly three big obstacles. They are i) reorganization of the NIRS, ii) a maternity leave of Core Leader F, and iii) moving to a new affiliation of members, including Dr. Jaks, the important member of the core. Especially, reorganization of NIRS and merging to QST resulted in drastic change of Core Leader J's team. All members, who were supposed to be core members in the plan and were indispensable for running experiments at HIMAC, left NIRS moving to new affiliations. Considering those circumstantial change, we had decided to do the followings. Firstly, we revised the experiment schedule and downsized the experiments. Secondly, we should recruit new members, not only in Japanese colleagues but also foreign researchers. Although the initial research plan has been significantly delayed and altered, the research activity continues and the international network has been established.

Experimental results

1. Wound healing of irradiated skin

It is well known that stem cells in "skin", which are involved in wound healing, are drastically reduced by irradiation. Comparison of an irradiated region to a non-irradiated region of the same sample is a good way to evaluate impact of irradiation on stem cell related biological responses, such as wound healing and carcinogenesis. To analyze phenomenon at the border of irradiated and non-irradiated tissue, photon irradiation has a large defect, such as diffraction. The shielded area is also exposed to photon irradiation, and

it is difficult to prevent the influence of radiation to skin, especially fair follicle stem cells, which are relatively radiosensitive. On the other hand, charged particle beams, including carbon ion beams, make it possible to clearly irradiate the boundary by taking advantage of the characteristics of charged particles. Therefore, we have used carbon beams for partial irradiation to investigate the difference in wound healing ability between the irradiated region and the non-irradiated region. Irradiated mice were photographed in Fig. 1. If the skin stem cells have significant contribution for wound healing, the wound in non-irradiated area might be smaller than the one of irradiated area, then we will be able to use this model for evaluate long distance effects between a wound and stem cells in a non-irradiated area. To our complete astonishment, no difference was observed in healing speed of wounds in the irradiated region and the non-irradiated regions in either of the two mouse strains (Fig 1. Lower panel). We are currently comparing to find differences between the irradiated region and the non-irradiated region, such as stem cell distributions and pathological analysis.

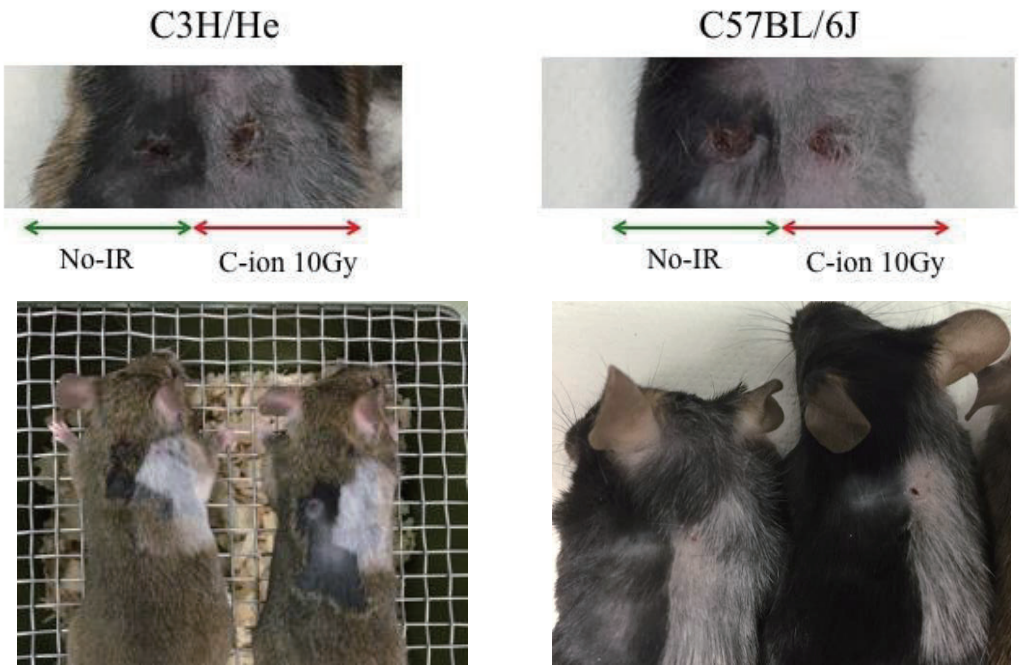


Fig. 1. Results of the wound healing experiments.

2. Effect of partial irradiation on tumor

Except the skin, partial irradiation on normal tissues is unrealistic since it is difficult to fix samples and precisely localize the irradiated area by using the current techniques. Therefore, we partially irradiated transplanted tumors as a model of stereoscopic organs. As a first step, we began to use the MRI which enabled us to make microstructure analysis. This is realized by a new collaboration with Dr. Aoki, who is specializing in the imaging techniques. We measured the time-course change in the same samples by using MRI (Fig. 2). Even though we irradiated those samples with high dose enough to kill 99.9% of the cancer cells *in vitro* condition, no clear alternations in the irradiated regions were observed up to now. We are going to investigate more details carefully and use additional contrast dyes to find any changes in the tumor after partial

irradiation.

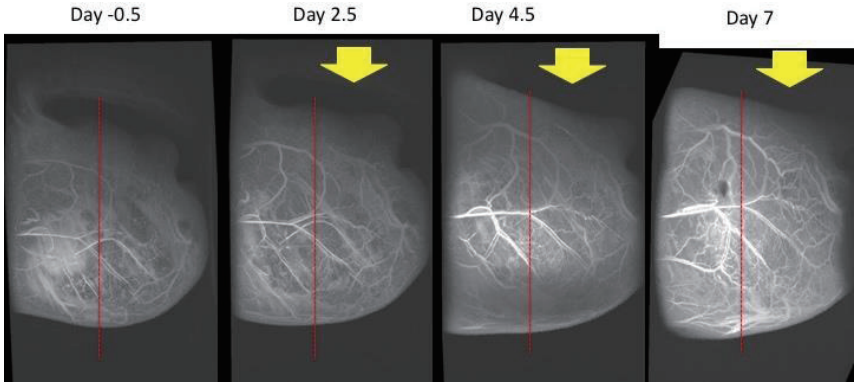


Fig. 2. MRI images of partial irradiated tumor.

As a next step, we observed details of pathological changes in the partially irradiated tumor. Partial irradiation on the transplanted tumor did not form clear boundaries between irradiated and non-irradiated regions (Fig.3. left panel), unlike irradiation on normal skin (Fig.3. right panel). Contrary to our expectation, cell death in the non-irradiated region and proliferating cell population in the irradiated region were observed (Fig.3. left-lower panel).

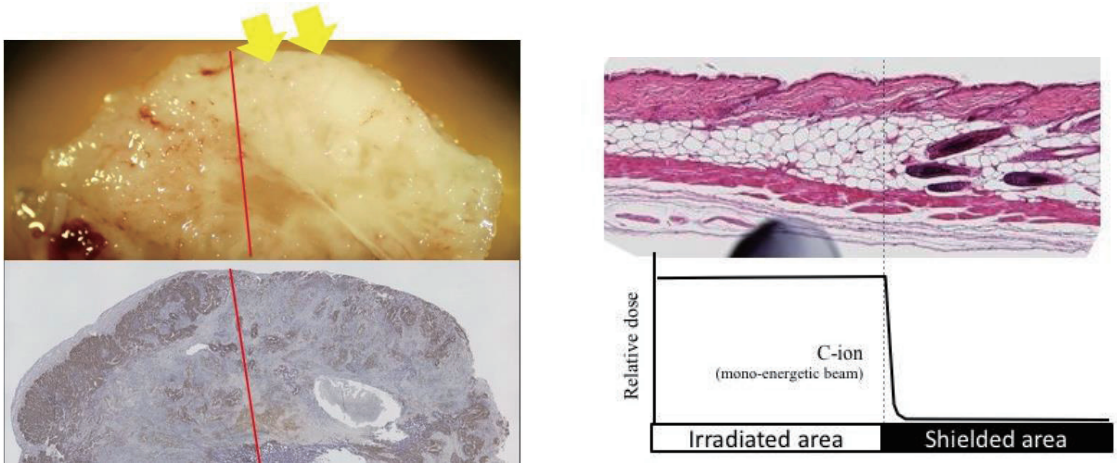


Fig. 3. Pathological analysis of partial irradiation.

3. New *in vitro* model (3D phantom)

In our initial studies using mouse models, we realized that mouse models have severe limitation about the analysis due to various reasons. Particularly, the influence of radiation induced cell death on the surrounding cells or non-irradiated cells, which are known as the bystander effect or the phoenix rising effect, are prevented the clear explanation of observed phenomena by presence of other cells, specially immune cells. As an alternative method, we began to use the 3D phantom, which was mainly developed by the Italian group. This phantom is a hydro gel using new materials and have several advantages for our project. It can easily construct any shapes with living cells and it has a clear body, which allow to observe inside by confocal microscopy. We have been evaluating the 3D phantom at NIRS, TIFPA and CIBIO.

Re-evaluation of the plan and expansion of the IOL collaboration

Summarizing above observation, it is clear that normal tissues and tumor showed completely different results. In addition, our results indicated that most of *in vivo* result might not be in line of our hypothesis based on in vitro data. One of the possible reasoning is that immune system may play an important role *in vivo* (and also tissue-specific) circumstances. Our original content might be able to rephrase as an investigation of "the influence (repair, second cancer *etc.*) of radiation-induced damage (*e.g.* cell death) on surrounding non-irradiated normal cells". However, under the premise of the initial plan, we have completely ignored the immune cells.

During the course of the above discussion, we exchanged the information with the Italian group. Since the Italian group and related researchers have had an interest in radiation-induced immune response, we decided to extend the collaboration network. Among the experiments performed with the Italian group, the additional immune-related experiments are analysis of the influence (anti/pro-tumor effects, inflammation) of radiation-induced damage via immune response" on surrounding non-irradiated normal and cancer cells.

This extended collaboration with the IOL team as a core, included researchers from USA and Germany other than Italians. In this collaboration, we have investigated molecular mechanisms of immune response by particle-beam irradiation. We presented the result of our collaboration at international conferences (PTCOG 2017, 2018, ERRS 2017), and two review articles were published, one of which was selected Top 10% in the immune research field, and one more was submitted.

In addition, we further develop the project/international collaboration to control the influence of "immune response-mediated " radiation effects.

Setting of export and import procedures

In this study, some of the specific analyses, such as pathological analysis, and experiments, including C-ion or proton irradiations, are able to achieve by a particular person or at the particular facility. Therefore, it was necessary to transport samples and materials frequently between several countries, international exports and imports. At present, the export control is very strict and the procedure is very complicated, including Japan. Therefore, we consulted with the NIRS administrative office, and we established the simplified procedure for exporting samples. Other researchers at NIRS can use the same simplified procedure when they practice the international collaboration.

Activities

1) Mutual visits and collaboration:

Dr. W. Tinganelli (TIFPA, Italy) and Dr. A. Helm (GSI, Germany) visited NIRS for discussions for initiating a new collaboration and preliminary experiments at HIMAC. 2016/2/13-2/25.

Dr. Shimokawa visited Karolinska Institutet, Stockholm, Sweden in attend the meetings for initiation and re-arrangement the IOL project with Dr. M. Kasper, Dr. J. Viljar (Estonia), Prof. Rune Toftgård, Prof. Peter Zaphiropoulos, Dr. Stephan Teglund, Prof. Staffan Strömblad, Dr. Casba Fint, Dr. Xiaoyan Sun and Dr. Ulrica Toster. 2016/2/25-3/3.
He also contributed to PhD defense (Anja Füllgrabe) and had a technical meeting for supervising of Miao Zhao and Rahman Mohammed Ferdous-ur, who are Ph.D. students.

Dr. Shimokawa visited Trento Institute for Fundamentals Physics Applications, Trento, Italy. Seminars at TIFPA and Santa Chiara Hospital. 2016/11/4-11/11.
He had meetings with Dr. W. Tinganelli, Dr. A. Helm, Dr. W. Bonani for exchanging progress reports of each groups and discussion to improve the 3D phantom system.
He also visits CIBIO (Dr. W. Bonani's Lab and animal rearing facility) and Proton center in Trento.

Dr. W. Tinganelli (TIFPA, Italy) and Dr. A. Helm (TIFPA, Italy) visited NIRS for discussion and preliminary experiments of radiation-induced immune response, and presentation in the IOL symposium 2017. They had a meeting with Dr. I. Aoki of NIRS for future collaboration, and attended a High-LET seminar. 2017/2/22-3/11.

Dr. V. Jaks (Tart University Hospital, Estonia) visited NIRS for discussion and preliminary experiments of the skin wound healing and presentation in the IOL symposium 2017. 2017/2/26-3/2.

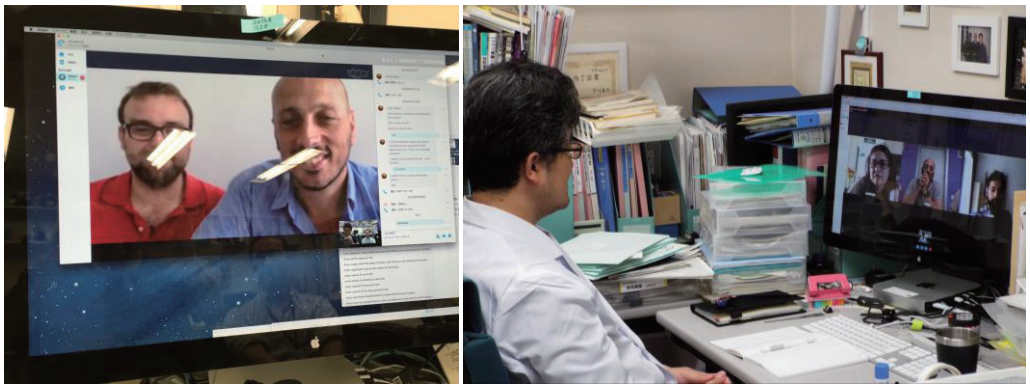
Dr. W. Tinganelli (TIFPA, Italy) and Dr. A. Helm (TIFPA, Italy) visited NIRS for discussions and experiments for 3D phantom and radiation-induced immune response, and presented in the IOL symposium 2018. 2018/6/10-6/28.

Dr. Shimokawa will visit Trento Institute for Fundamentals Physics Applications, Trento, Italy for discussion and experiments for radiation-induced immune response under a space travel condition. 2018 Autumn.

Dr. Shimokawa will visit University of Tartu, Estonia for giving seminars for medical students and Ph.D. students and discussions for further collaboration with Dr. V. Jaks. 2018 Autumn.

Web meetings:

More than 25 times of web meeting with multi locations.



Publications

- Ebner DK, Tinganelli W, Helm A, Bisio A, Palma S, Francesco N, Yamada S, Kamada T, Shimokawa T, Durante M. Generating and grading the Abscopal Effect: Proposal for comprehensive evaluation of combination immunoradiotherapy in mouse models. *Transl Cancer Res*. 6: S900-913, 2017.
- Ebner DK, Tinganelli W, Helm A, Bisio A, Yamada S, Kamada T, Shimokawa T, Durante M. The Immunoregulatory Potential of Particle Radiation in Cancer Therapy. *Front Immunol*.8:99, 2017. (selected as a top 10% articles)

External grant

- Grant-in-Aid for Exploratory Research, JSPS 2015.4-2017.3, Takashi Shimokawa
- Chairman's Fund, QST, 2017.8-, Takashi Shimokawa

Submitted grant applications

1. Melanoma Research Alliance, 2017 (not adopted)
2. Grant-in-Aid for Scientific Research (B), 2017(not adopted)
3. Grant-in-Aid for Scientific Research (B), 2018 (not adopted)
4. The Harry J. Lloyd Charitable Trustee, 2018 (waiting)

Whole Gamma Imaging (WGI) Core

CORE LEADER (J)

Taiga Yamaya, Ph. D., Team Leader of Imaging Physics, NIRS, QST
yamaya.taiga@qst.go.jp
Research interest: development of next generation positron emission tomography (PET) instrumentations as well as new detectors and image reconstruction algorithms

CORE LEADER (F)

Katia Parodi, Ph. D., Professor, Chair of Experimental Physics - Medical Physics,
the Ludwig-Maximilians-Universität in Munich (LMU)
Katia.Parodi@physik.uni-muenchen.de
Research interest: high precision image-guided radiotherapy with a special focus on ion beams, from advanced computational modeling to experimental developments and clinical evaluation of novel methods for in-vivo ion range monitoring

Core Members

Eiji Yoshida	(QST/NIRS)	Peter Thirolf	(LMU)
Fumihiko Nishikido	(QST/NIRS)	Georges Dedes	(LMU)
Hideaki Tashima	(QST/NIRS)	Guillaume Landry	(LMU)
Akram Mohammadi	(QST/NIRS)	Ingrid Valencia Lozano	(LMU)
Sodai Takyu	(QST/NIRS)	Silvia Liprandi	(LMU)
Go Akamatsu	(QST/NIRS)	Vasiliki Anagnostatou	(LMU)
Atsushi Tsuji	(QST/NIRS)	Mohammed Safari	(LMU)
Kotaro Nagatsu	(QST/NIRS)	Maximilian Grosch	(LMU)
		Maria Kawula	(LMU)
		Tim Binder	(LMU)
		Rita Viegas	(LMU)

Purpose of the Project:

Positron emission tomography (PET) is recognized as a successful method to pursue cancer diagnosis and molecular imaging. However, for further improvement regarding imaging of lower activity concentrations and a wider spectrum of radionuclides, we need to break through the principle of PET itself. In this project, we propose a new concept of whole gamma imaging (WGI), which is a novel combination of PET and Compton imaging. For positron emitters, missing pairs of annihilation photons, at least one of which is undetected, can be used for imaging. In addition, further impact can be expected for triple gamma emitters such as ^{44}Sc, that emits a pair of 511 keV photons and a 1157 keV gamma almost at the same time. In

theory, localization from a single decay might be possible by identifying the intersection point between a coincidence line and a Compton cone. In this project, we aim at the world's-first realization of WGI by merging our potential detector technologies, depth-of-interaction (DOI) detector technologies at NIRS and Compton camera technologies at LMU.

Summary of the Project

Our research activities in three years are summarized as below:

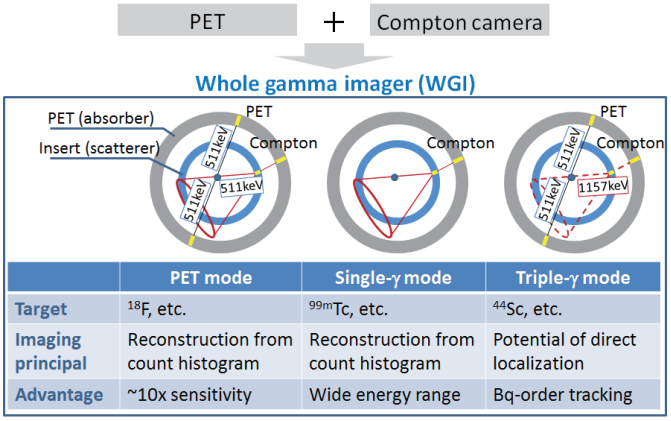
- Well-funded in each core to realize a concept of WGI
- Progress in detector development by mixing novel technologies of DOI (NIRS) and Compton (LMU)
- Success in the world's first realization of WGI to show a proof-of-concept
- Active researchers/students exchange between NIRS and LMU

(1) Concept of WGI

WGI is a concept that utilizes all detectable gamma rays for imaging (Fig. 1). An additional detector ring, which is used as the scatterer, is inserted in the field of view of a conventional PET ring so that single gamma rays can be detected by the Compton imaging method.

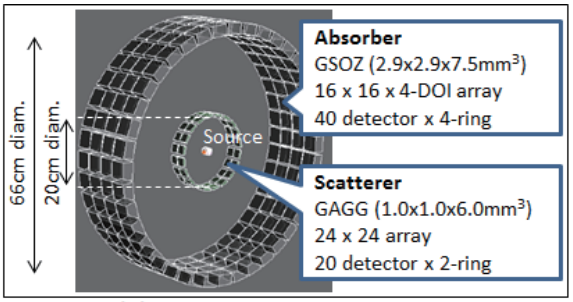
(2) Simulation

Using GEANT4, we simulated an "insert geometry", where a scatter ring (24 x 24 array of 1 x 1 x 6 mm^3 GAGG crystals, 20 cm diameter and 5 cm long,) was inserted into a PET ring (16 x 16 x 4-DOI array of 2.9 x 2.9 x 7.5 mm^3 GSOZ crystals, 66 cm diameter and 22 cm long) as shown in Fig. 2 (a). We simulated a 511keV source for performance evaluation of Compton imaging. We also simulated a ^{22}Na point source as a source of triple gamma emitter (e+ and

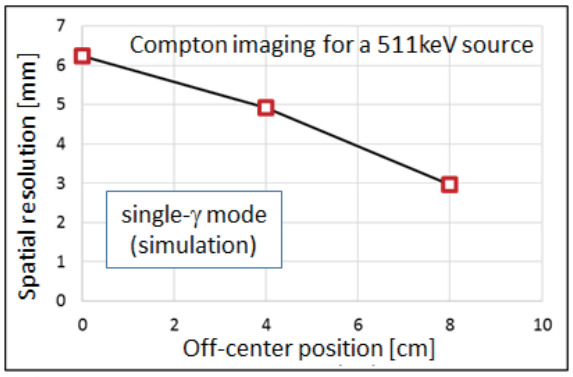


	PET mode	Single-γ mode	Triple-γ mode
Target	^{18}F, etc.	^{99m}Tc, etc.	^{44}Sc, etc.
Imaging principal	Reconstruction from count histogram	Reconstruction from count histogram	Potential of direct localization
Advantage	~10x sensitivity	Wide energy range	Bq-order tracking

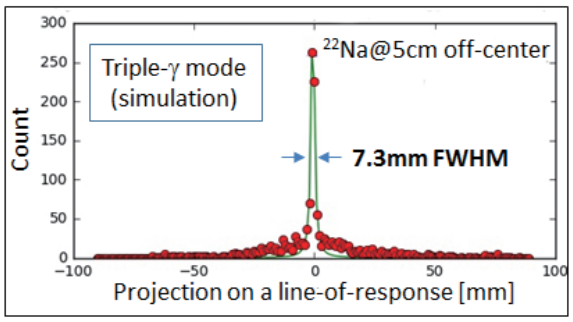
Fig. 1 Concept of the WGI system, which has three modes of operation.



(a) Simulated WGI geometry



(b) Image reconstruction for the single-γ mode



(c) Simple projection for the 3γ mode

Fig. 2 Illustration of the simulated WGI (a) and simulation results: spatial resolution of the single-γ mode (b) and localization performance of the triple-γ mode (c).

1274 keV gamma ray).

In the single-γ mode, spatial resolution for the 511 keV source obtained by 3D list-mode OSEM was 6.2 mm FWHM (center) – 3.0 mm FWHM (8 cm off-center) (Fig. 2 (b)). In the triple-γ mode, the source position distribution projected on a LOR was 7.3 mm FWHM at the 5 cm off-center position without applying any image reconstruction (Fig. 2 (c)). This localization performance was much greater than the state-of-the-art PET technology of time-of-flight measurement, which gives around 6.0cm - 7.5cm localization.

(3) Experimental characterization of detector components for Compton imaging

In addition to the well-established DOI GSOZ detectors, which was modeled in the simulation, we pursued further development and characterization of monolithic detector crystals ($LaBr_3$ and $CeBr_3$) of excellent spatial and temporal resolution as well as Si strip detectors, which serve as components of a Compton camera system being realized at LMU. First comparative studies were also performed among these different detector solutions, along with GAGG arrays, in order to confirm the technology for the first WGI prototype and identify possible solutions for its future optimization.

Monolithic detector characterization

We realized a dedicated laboratory set-up, which enables automated scanning of the front surface of the considered monolithic crystals with tightly collimated ^{60}Co and ^{137}Cs sources in 10^4 different positions. These data serve as reference 2D library for position reconstruction using a "Categorical Average Pattern" algorithm (figure 3), which confirmed the possibility to achieve 2.9 mm and 4.8 mm resolution at the considered photon energies of. 1.3 MeV and 662 keV, respectively.

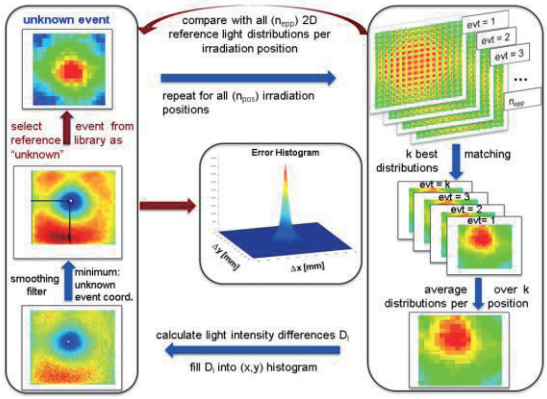


Fig. 3 Workflow for extracting the interaction position of an unknown impinging γ-ray in the monolithic $LaBr_3$(Ce) scintillator, in this case taken from a reference library to quantify the spatial resolution from the FWHM (Full Width Half Maximum) of the error histogram created from deviations between real and calculated photon interaction positions.

Combination of LMU and QST/NIRS detector technologies for Compton imaging

We adapted the data readout systems of detectors available at both institutions to enable different combinations of scatterer and absorber for Compton imaging. Point sources were imaged using the 22 × 22 array of 0.9 × 0.9 × 6 mm^3 GAGG scatterer developed at QST/NIRS for the first WGI prototype, in combination with either a 50 × 50 × 30 mm^3 monolithic $LaBr_3$ (or $CeBr_3$) or a pixelated DOI detector of segmented (32 × 32 × 4-layer array of 1.46 x 1.46 x 4.5 mm^3) LYSO as absorber. The results for different source-absorber distances (from 50 to 200 mm), for a fixed 45 mm distance between source and scatterer, confirmed the imaging capability of all setups (figure 4), with increased spatial resolution (at the cost of

decreased sensitivity) with increasing source-absorber distance. A direct comparison of all settings is ongoing and will be published soon.

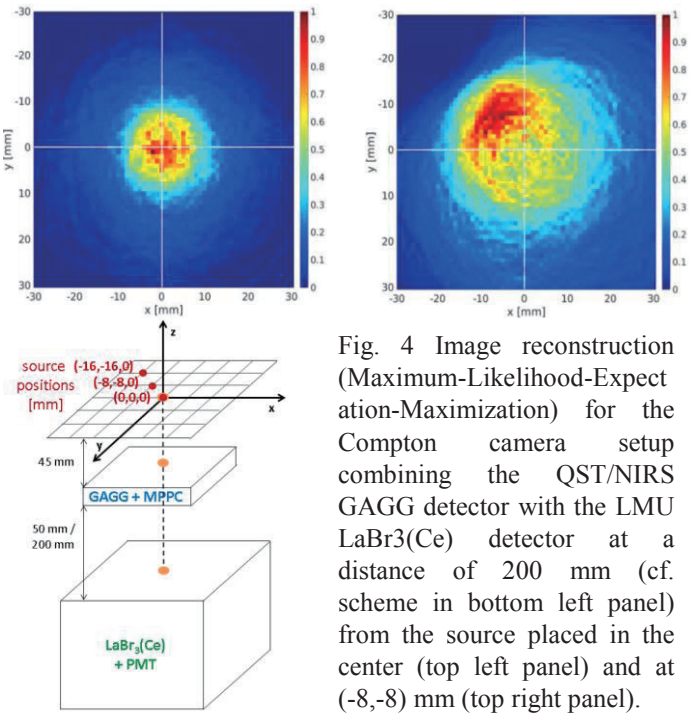


Fig. 4 Image reconstruction (Maximum-Likelihood-Expectation-Maximization) for the Compton camera setup combining the QST/NIRS GAGG detector with the LMU LaBr3(Ce) detector at a distance of 200 mm (cf. scheme in bottom left panel) from the source placed in the center (top left panel) and at (-8,-8) mm (top right panel).

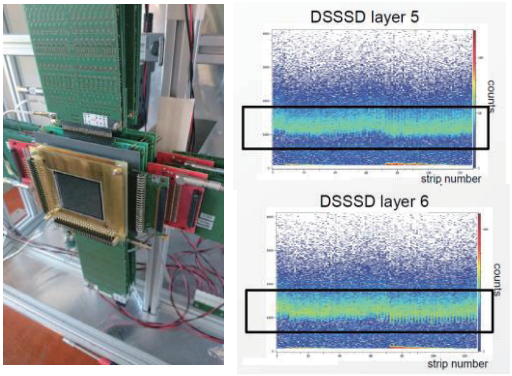


Fig. 5 Experimental assembly (left) and acquired signal (right) in the last two layers of thin (0.5 mm) double-sided Si-strip detectors exposed to energetic (~4-6 MeV) γ-rays produced in proton-induced nuclear interactions. Signals originated from Compton scattered electrons are clearly visible in the black rectangular box.

Investigating a Si-based scatterer for WGI

Although GAGG can be considered a very good compromise solution as Compton scatterer, owing to its high effective atomic number and moderate energy resolution, Si could offer an even better solution for a next generation WGI. Hence, we thoroughly characterized and very recently upgraded the readout electronics of silicon strip detectors under development at LMU, featuring both 0.5 mm and 1 mm thick modules, which might in future be combined for optimal detector thickness for WGI. An example of promising first detector characterization during exposure to energetic (4-6 MeV) γ-rays is shown in figure 5.

(4) Prototype WGI development and experiment

We developed the first prototype of the WGI system based on well proven technologies (Fig. 6). The major

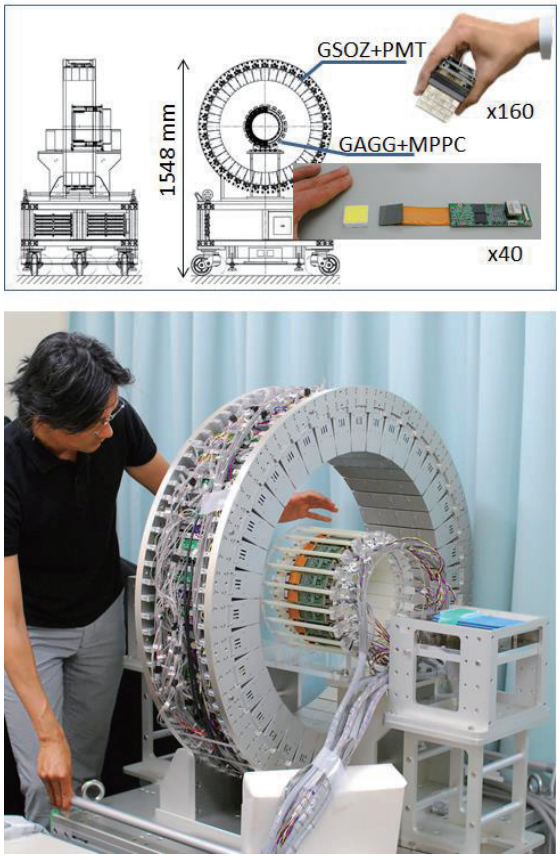


Fig. 6 Developed prototype WGI system.

parameters of the prototype are almost the same as those in the simulation. All interaction events were recorded as list-mode data, and event selection such as coincidence detection was done in software. In the single-γ mode, spatial resolution for the ^{137}Cs point source obtained by 3D list-mode OSEM was 12.4 mm FWHM (center) - 5.9 mm FWHM (8 cm off-center).

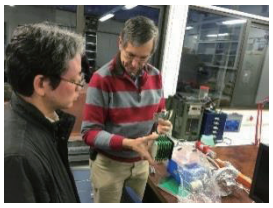
(5) Discussion and conclusion

Simulation and experimental results showed the initial feasibility of the WGI concept. In the prototype, the results of the coincidence detection suggested that the scatterer ring requires finer tuning, while the absorber ring works well. This finer tuning may improve the performance of the single-γ mode, in which differences were almost twice as large between simulation and experiment results. Moreover, other detector technologies for Compton imaging at different level of maturity have been investigated, and we have built the premises for direct experimental comparison of different detector solutions, thus promising future optimization of WGI.

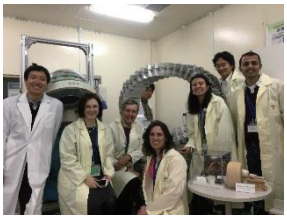
Activities

(1) Personnel exchanges and activities of experiments

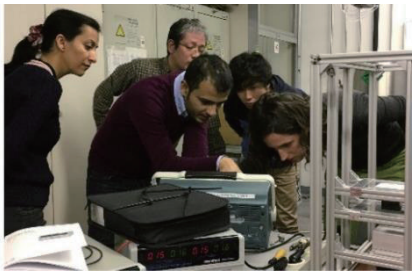
First visit to LMU was made by three NIRS researchers (Yamaya, Yoshida and Nishikido) in November 2015.



Then five researchers from LMU (Parodi, Thirolf, Aldawood, Lozano and Liprandi) visited NIRS to learn the DOI detector technology in March 2016.



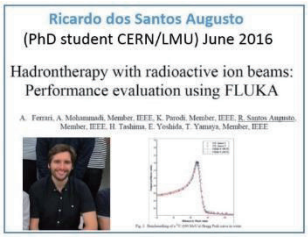
The next visit to LMU to explore the best combination of detector elements was made by four NIRS researchers (Yamaya, Nishikido, Mohammadi and Takyu) in January 2017.

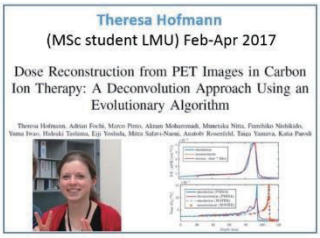


The improved detector module followed by the experimental results was brought to LMU and tested again by three NIRS researchers (Yamaya, Nishikido and Takyu) in January 2018.

In addition to these collaborative experiments, NIRS researchers have visited LMU to have meetings on May 20, 2016 (Yamaya), June 19, 2017 (Yamaya) and February 27 2018 (Yamaya and Akamatsu).

Yamaya-lab has accepted two internship students from Parodi-lab, Augusto (Ph. D student of CERN/LMU) in June 2016 and Hofmann (MSc student of LMU) from February until April 2017.





Presentations

[1] H. Tashima, C. Kurz, E. Yoshida, J. Debus, K. Parodi, T. Yamaya, "Patient Data-Based Monte Carlo Simulation of in-Beam Single-Ring OpenPET Imaging," 2015 IEEE Nuclear Science Symposium and Medical Imaging Conference, M5A1-4, 2015 (oral, 2015/11/6, San Diego).

[2] E. Yoshida, H. Tashima, C. S. Levin, K. Parodi, T. Yamaya, "Simulation Study of a DOI-Based PET-Compton Imaging System for Positron Emitters," 2015 IEEE Nuclear Science Symposium and Medical Imaging Conference, M4CP-2, 2015 (poster, 2015/11/5, San Diego).

[3] E. Yoshida, H. Tashima, C. S. Levin, K.Parodi, T. Yamaya, "Sensitivity and spatial resolution simulation of a PET-Compton insert imaging system," 2016 IEEE Nuclear Science Symposium and Medical Imaging Conference, M04D-10, 2016. (poster, 2016/11/2, Strasbourg)

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[5] T. Yamaya, E. Yoshida, H. Tashima, A. Tsuji, K. Nagatsu, M. Yamaguchi, N. Kawachi, Y. Okumura, M. Suga, K. Parodi, "Whole gamma imaging (WGI) concept: simulation study of triple-gamma imaging," J. Nucl. Med., vol. 58, no. supplement 1, 152, 2017 (SNMMI 2017 Annual Meeting, oral, No. 152, 2017/6/12, Denver, Highlighted)

[6] T. Yamaya, E. Yoshida, K. Nagatsu, H. Tashima, Y. Okumura, M. Suga, N. Kawachi, K. Kamada, P. G. Thirolf, K. Parodi, "Whole gamma imaging (WGI) concept: demonstration of 44Sc triple gamma imaging," 2017 World Molecular Imaging Congress Program Schedule and Abstract Book, LBA 24, 2017. (oral, 2017/9/14, Pennsylvania Convention Center, Philadelphia)

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[8] E. Yoshida, H. Tashima, Y. Okumura, M. Suga, N. Kawachi, K. Kamada, K. Parodi, T. Yamaya, "Concrete realization of the whole gamma imaging concept," 2017 IEEE Nuclear Science Symposium and Medical Imaging Conference, M-21-2, 2017. (2017/10/28, oral, Atlanta)

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Publications

[1] E. Yoshida, H. Tashima, C. S. Levin, K. Parodi, T. Yamaya, "Simulation Study of a DOI-Based PET-Compton Imaging System for Positron Emitters," Conf. Rec. 2015 IEEE Nuclear Science Symposium and Medical Imaging Conference, M4CP-2, 2016.

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[3] H. Tashima, Christopher Kurz, E. Yoshida, W. Chen, J. Bauer, J. Debus, K. Parodi, T. Yamaya,

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[6] T. Yamaya, E. Yoshida, H. Tashima, Y. Okumura, M. Suga, N. Kawachi, K. Kamada, K. Parodi, "Concrete realization of the whole gamma imaging concept," Conf. Rec. for 2017 IEEE Nuclear Science Symposium and Medical Imaging Conference, M-21-2, 2018. (to be published)

[7] R.S. Augusto, A. Mohammadi, H. Tashima, E. Yoshida, T. Yamaya, A. Ferrari, K. Parodi, "Hadrontherapy with radioactive ion beams: Performance evaluation using FLUKA," to be submitted to Phys. Med. Biol.

[8] T. Hofmann, M. Pinto, A. Fochi, A. Mohammadi, M..Nitta, F. Nishikido, Y. Iwao, H. Tashima, E. Yoshida, M. Safavi-Naeini, A. Chacon, A. Rosenfeld, T. Yamaya, K. Parodi, "Dose reconstruction from PET images in carbon ion therapy: a deconvolution approach,", to be submitted to Phys. Med. Biol.

External grants

Relating funding at NIRS

PI	Grant	Duration	Direct expense
Yamaya	Kakenhi Kiban A	4/2016-3/2019 (3y)	¥33,600k
Yoshida	Kakenhi Kian C	4/2015-3/2018 (3y)	¥3,600k
Mohammadi	Kakenhi Kian C	4/2017-3/2020 (3y)	¥3,700k
Yamaya	QST President's Fund	4/2016-3/2020 (4y)	¥17,000k (excl. FY2019)

Relating funding at LMU

PI	Grant	Duration	Direct expense
Parodi/ Anagnostatou	EU MSCA IF	06/2017-05/2019	170 kEUR
Parodi/Safari	Humboldt Foundation	02/2018-01/2020	~150 kEUR
Parodi	EU ERC CoG	11/2017-10/2021	1,600 kEUR
Parodi/Thirolf	DFG	10/2012-12/2018	1,200 kEUR
Thirolf	BFS	02/2018-01/2021	263.5 kEUR

Awards

[1] Hideaki Tashima, Japanese Society of Medical Physics, the President Award, 2016/4/14

[2] Taiga Yamaya, Japanese Society of Medical Physics, the President Award, 2017/4/14

[3] Theresa Hofmann, 3rd place at the LMU's first "Physics Student Research Conference", 2017/6

[4] Silvia Liprandi, Top-10 position in the (2-staged) Student Competition at the Annual Meeting of the German Society for Biomedical Engineering, 2017/9

3．第3期IOLシンポジウム

IOL Symposium 2017

International Open Laboratory Symposium 2017

Date : 28 Feb, 2017 13:00-18:00

The International Open Laboratory (IOL) of NIRS began in 2008 in order to encourage collaboration with overseas researchers. Continuing the successful 1st and 2nd term, the 3rd term of IOL began in 2015 with 4 groups, named "research cores". The International Open Laboratory Symposium 2017 will be held on February 28, 2017, and the activities and status of the 4 cores will be reported.

Venue Lecture Hall, 2nd floor of Research Building for Charged Particle Therapy
National Institute of Radiological Sciences
National Institutes for Quantum and Radiological Science and Technology
Anagawa 4-9-1, Inage-ku, Chiba
http://www.qst.go.jp

Registration No advance registration, no registration fee

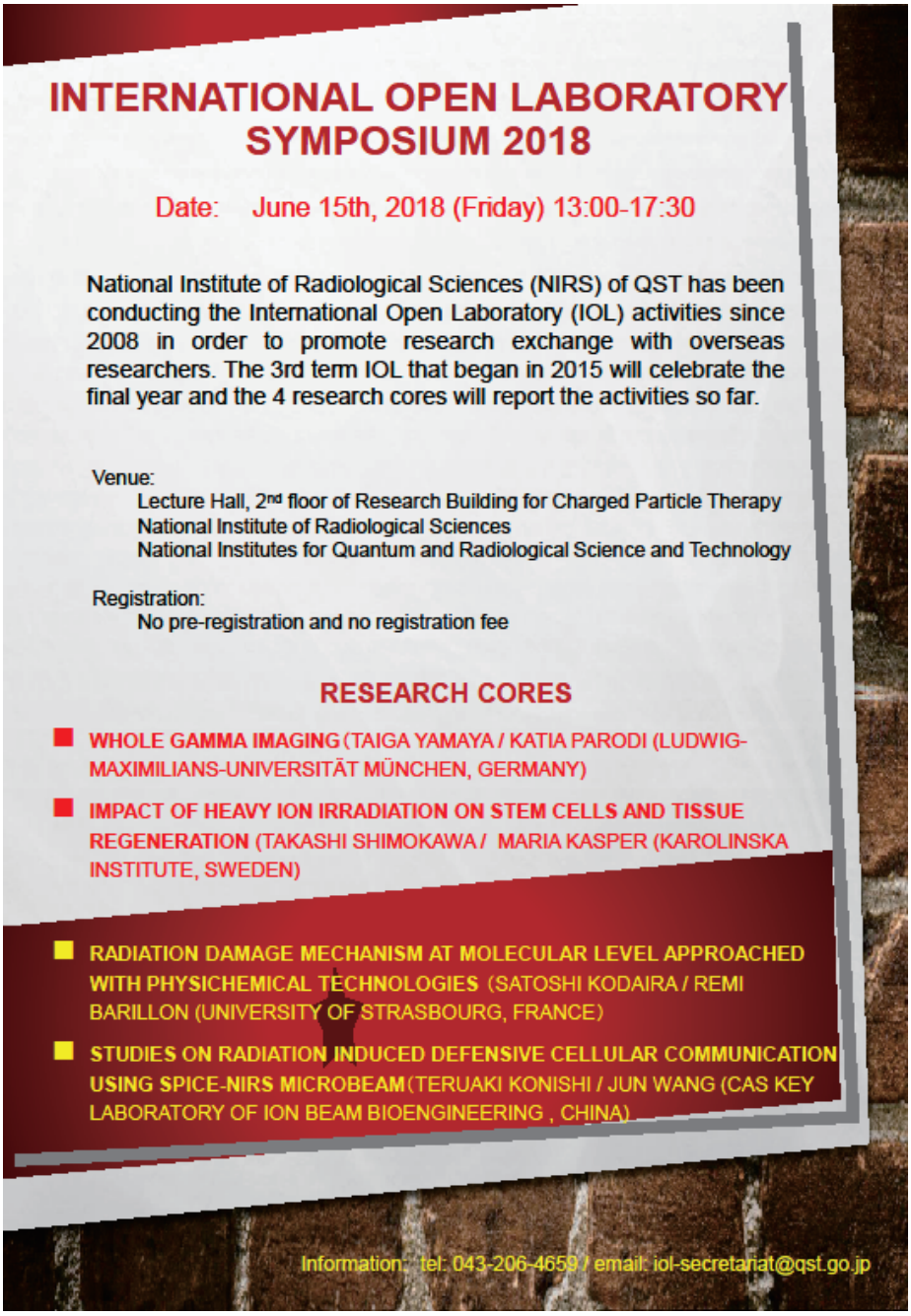
Program (tentative)

- ◆ Report of Research Cores
  - ◇ Whole gamma imaging
    - Taiga Yamaya
    - Katia Parodi (Ludwig-Maximilians-Universität München, Germany)
  - ◇ Impact of heavy ion irradiation on stem cells and tissue regeneration
    - Takashi Shimokawa
    - Maria Kasper (Karolinska Institute, Sweden)
  - ◇ Radiation damage mechanism at molecular level approached with physiochemical technologies
    - Satoshi Kodaira
    - Remi Barillon (University of Strasbourg, France)
  - ◇ Studies on radiation induced defensive cellular communication using SPICE-NIRS microbeam
    - Teruaki Konishi
    - Jun Wang (CAS Key Laboratory of Ion Beam Bio-engineering , China)
- ◆ Other examples of international collaboration
- ◆ Expectations and comments

Information E-mail: iol-secretariat@qst.go.jp QST

IOL Symposium 2017

IOL Symposium 2018



IOL Symposium 2018

４．第３期 IOL　予算執行実績

第3期IOL　予算執行実績

	1年目（2015-4/1～2016-8/31）	2年目(2016-9/1～2017-8/31)	3年目(2017-9/1～2018-8/31)
4コア	6,473,249	4,044,530	3,472,387
事務局	136,504	1,190,559	1,835,933
合計	**6,609,753**	**5,235,089**	**5,308,320**