諾率等を紹介する。

研究計画策定まで

2008年に組織された垣添班のメンバーに筆者らの一人が選定され，胸部CT検診の無作為化比較試験計画を策定することになったため，肺がん検診の専門家，疫学者，数理統計学者などの協力を得て計画を作成した。詳細は別稿 (8) に譲るが，概略を示すと，

1. 50-64歳の男女に対して，10年間の胸部CTが胸部X線に比べて40%の肺がん死亡減少効果があると見込み，妥当な応諾率，コンタミネーション (対照群に割付されたにもかかわらず他の医療機関などで胸部CTを受診してしまうこと) を設定すると，参加者は50000人が必要
2. 対照群では現行検診を10年間，研究群では喫煙者では低線量CT検診 (＋喀痰) を10回，非喫煙者では低線量CT検診は1，3，7年目の3回で残りは現行検診
3. X線とCTを比較する必要があるため対照群のX線受診率も高い必要があることから，それが見込める「ある年度の肺がん検診受診者」を対象とする
4. 検診費用は，同様の検査での健康保険収載の約90%とし，説明や事務的費用・追跡費用も含む。その結果，研究費用は15年で35億円超，年間2億円超となった
5. 同時に「CT測定による内臓脂肪と生活習慣病に関する大規模前向きコホート研究 (採血・腹部内臓脂肪のCTによる測定などとその後の各種疾患発症との関連を探るコホート研究)」を策定し，胸部CT検診のRCTで「対照群」となった人は参加することもできる，というオプションを提示する

この研究計画を金沢医科大学倫理委員会に提出し，2009年9月28日に承認された (受付番号No.91)。

パイロットスタディの計画とその目的

予算規模が大きいため「戦略研究」などでの採用を期待したが実現には至らなかった。そこで，パイロットスタディとして，地元との話し合いが進んでいた石川県と岡山県の一部の市町で先行して開始することになった。

このパイロットスタディの初年度の目的は，①研究全体の流れにおける多種の書類・ツールを作成する，②研究の実際における問題点を明らかにして計画を改善する，③対象者の何割が研究に参加するかを把握する，の3点であった。次年度以降では，④次年度以降のコンプライアンスとコンタミネーションがどの程度かを把握する，⑤精密検査結果を把握し，その適切性を評価すると共に精密検査システムの整備を行う，⑥検診受診により惹起される参加者の「不安感」の変化を検討する，などの目的も設定している。

参加自治体の選定

石川県では，2009年春に県内全市町村に対して，胸部CT検診の無作為化比較試験計画への参加希望に関するアンケートを行った。その際に希望のあった4市町村のうち「羽咋市」を第1候補として，市に対して研究計画を説明した結果，羽咋市はこの研究および内臓脂肪コホート研究に参加することになった。

検診日・場所の決定と共に，本年の肺がん検診終了直後に対象者の選定を行い (社会保険加入者は市町村で十分情報を把握していないため除外)，郵送で参加を募り，希望者に対して改めて受診の日時を通知する，などの段取りを決定し，以下のような書類・ツールの作成を行った。

必要な書類・ツールの作成

1. 説明会用に約17分間のインタビュー形式の説明ビデオを作成した。CT肺がん検診の効果は未確定であること・ランダマイズ・不利益・途中で研究中止となることもあること・などに関しても十分に説明した。
2. 説明用文書の作成を行った (図1)。「事前の郵送」「説明会での資料」の両用に使用できるような形式で，かつ「説明・同意文書」の内容を盛り込んだものにした。
3. 日時・場所等を見やすくした「研究参加勧奨チラシ」を作成した。
4. 仮参加申込書を作成した (図2)。「目的」「方法 (ランダマイズ)」「追跡調査」などの11項目を理解していること，適格性，および参加希望時間を確認できる書式とした。改訂版では事前のランダマイズが可能なように喫煙歴を追加した。
5. 研究参加同意書兼問診票を作成した。仮参加申込書の内容に加え，追跡調査用の個人情報，住民検診以外の検診受診，がん罹患などの情報を追加した。
6. 個人情報，問診票の内容，検診結果などを一元管理できるデータベースを作成した。
7. 羽咋市の封筒と羽咋市健康福祉課課長名での添え書きを用意してもらった。
8. 検診受診者の意識調査および不安度調査の目的で，「健康関連QOL尺度SF8の8項目」「HADS日本語版14項目のうち5，7，9，14の4項目」「CT検診に関する知識とイメージを問う3項目 (我々が作成)」のアンケートを作成した。
9. 説明文書および説明会での説明内容の理解度調査のアンケートを作成した。
10. 説明会当日の流れを説明したチラシを作成した。
11. 当日不参加を決定した人のために不参加理由書を作成した。

実際のリクルート業務の流れ

1. 本年の肺がん検診受診者のうち，2011年4月1日現在で50-64歳の男女で国民健康保険加入者のリストを作成した。
2. その全員に対して，①市からの添え書き，②研究参加勧奨チラシ，③胸部CTによる肺がん検診の無作為化比較試験の説明書，④仮参加申込書，⑤返信用封筒，を，⑥羽咋市の封筒に入れて郵送した。

厚生労働省壇添選胸部CT検診小班（小班長：金沢医科大学教授　佐川元保）

肺がん検診研究プロジェクトの説明書

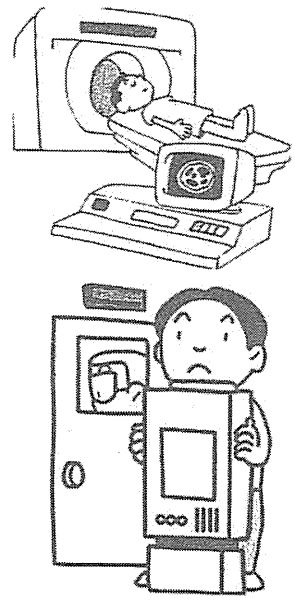
研究名：低線量CTによる肺がん検診の精度および死亡減少効果評価のための個人単位ランダム化比較試験

今なら無料で精密な検診が受けられます

なぜ、このような検診の研究が企画されたのですか？

肺がんによる死亡は増加しており、その対策は国家的にも重要です．現在日本で行われている胸部X線検査と喀痰細胞診検査は肺がんによる死亡を減らす効果があることがわかっておりますが万能ではなく、検診を受けても肺がんで亡くなる患者さんもいるのが実情です。

最近，胸部のCT検査（コンピューター断層撮影）を肺がん検診に用いる方法が一部で行われ始めました．その結果，多くの早期がんが見つかるようになったのですが，一方で，本来ならば治療する必要のないような病変も手術してしまったりする例があるのではないか，ということも危惧されており，CT検診とX線検診のどちらがより有益であるかはわかっていません．CT検診とX線検診を比べるために厚生労働省の研究班でこの研究が計画されました。全国で行う計画を立てましたが、先駆けとして全国で２つの市町が選ばれ、その一つが羽咋市です。

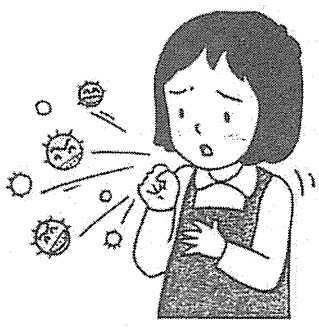


どのように行われるのですか？

CT検診は小さなものも見つかるかわり余計なものも見つけるかもしれず、X線検診より良いのかは現在のところ不明です。そのため、どちらかが「損」だの「得」だのということはありません．この研究では「CTとX線のどちらの検査法でも良い」と言っていただける方を集め，コンピューターで公平に分け、半分の方にCT、もう半分の方にX線検診を行っていただきます．

X線とCTのどちらの検査も、肺がんを数多く診断・治療している私たち専門医が、検査したフィルムを責任をもって診断します。

タバコをたくさん吸っていた方の場合は、早期がんを見つけるためには痰の検査も必要ですので、X線とCTのどちらの検査になっても、痰の検査も行います。



来年以降はどうなるのですか？

１－２年では効果が不十分ですので、この研究は10年間行う予定です。X線の方はX線を1年に1回、計10回受けることになります。一方、CTの方はタバコを吸っていたかどうかで回数が変わります。タバコを吸っていた方はCTを年に1回10年間ですが、タバコをあまり吸っていなかった方は10年間にCTを3回、X線を7回予定しています。この理由は、タバコを吸っていなかった方は3回程度で十分だろうと考えられているからです。逆にいえばタバコを吸っていた方はそれでは不十分だと考えられているのです。その点からも、なるべく早く禁煙することをお勧めします。

図1a, 1b, 1c. 低線量CTによる肺がん検診の無作為化比較試験の説明書 (原本はカラー)

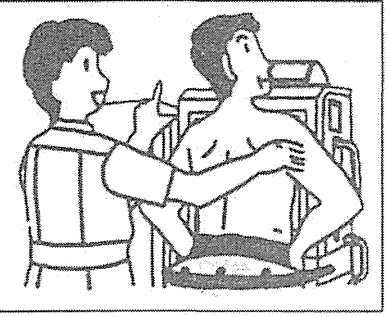
実際に参加するにはどうしたら良いのですか？

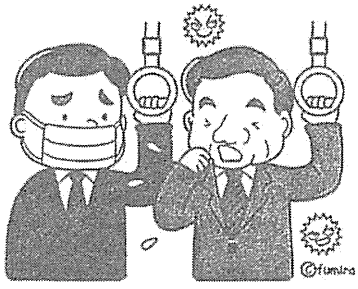
この研究に参加するには、この説明書を良く読んでいただき、内容を理解して参加しようと思われたら、同封の「参加（仮）申込書」に必要事項を書いていただき、10月に行われる3回の説明会のうち出席希望の日と午前か午後かを丸で囲んで、返信用封筒でご返送ください。

事務局であなたが参加可能であることを確認できましたら、参加決定日を記載した「参加（仮）確認書」を郵送いたします。その参加決定日に会場に来ていただいて、その場でビデオによる説明、および口頭による説明を聞いていただき、納得されたら正式に参加となります。

その後、CT検診の方とX線検診の方にわかれることになりますが、初年度は今年の分のX線検診をつい先頃受診していますので、X線をもう一度撮ることはしませんので、それで終了です。ただし、「腹部内臓脂肪CT検査＋採血検査」をオプションとして無料で受けることもできますので、御希望の方は申し出てください。CT群の方は、そのまま胸部CTを撮影していただくことになります。結果は後日お知らせします。

来年度以降も、CT群の方もX線群の方も、通常の検診よりもさらに慎重に診断しますので、通常の検診とは実施日を変えて羽咋市体育館で行う予定です。参加者の方にはあらかじめ通知いたします。その際には、通常は肺がん検診と同時に行っている特定検診なども同時に行えるように手配しておきます。





費用はかかるのですか？　何年間行うのですか？

検診で行うX線検査、CT検査、痰の検査は、いずれも事務局が負担しますので、皆さんの負担はなく、無料になります。ただし、検診で異常が見つかって医療機関で精密検査を行う場合には、通常の保険診療として通常の窓口負担が生じます。

この研究は10年間行う予定ですが、国の予算で動いているため、事業仕分けなどで予算がおりなくなれば途中で中断する可能性もあります。

CT検診で予想される利益と不利益には何がありますか？　検診で必ずがんが早期に見つかるのですか？

胸部CT検診を受けた方の予想される利益としては，肺がんによる死亡をX線よりもさらに減らすことができるかもしれない，ということが挙げられます．一方，不利益としては，第一に放射線被曝の問題が挙げられますが，今回対象の年齢の方ではそれほど問題ありません．その他の不利益としては，治療の不要な良性病変のために精密検査や手術が必要になる可能性があります．また，がんであっても非常に増大速度が遅く天寿を全うできるようなものを手術してしまう可能性もあります．あまり小さなものまで精密検査を行うと受診する方の不利益になるため、この研究では日本ＣＴ検診学会の基準に従って「要精密検査」とするように規定しています。また，CTとX線のどちらの場合でも、精密検査や治療を行っている中で医療上のトラブル・合併症に巻き込まれる可能性は０にはできませんので、そのような可能性はあります．また、非常に小さながん、急速に増大するがん、見えにくい場所にできるがんは、検診では見つけられないことがあります。

説明会および検診の日時：
平成22年　10月8日（金）
10月15日（金）
10月24日（日）
いずれも受付は9—16時
場所：羽咋市役所横体育館

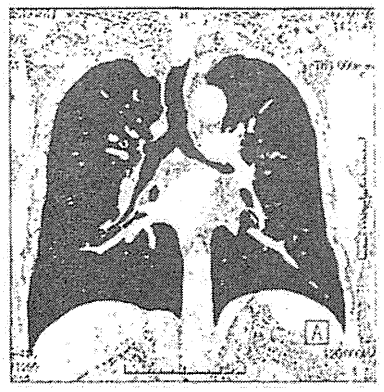


図1b.

参加するときの条件はありますか？

この研究に参加するためには、いくつかの条件があります。

1．本年の10月に3回ほど説明会を開きますので、そのいずれかの日程に出席のうえ説明を直接聞いていただき、参加の意思を確認する必要がありますので、どの日程にも出席できない人は参加できません。

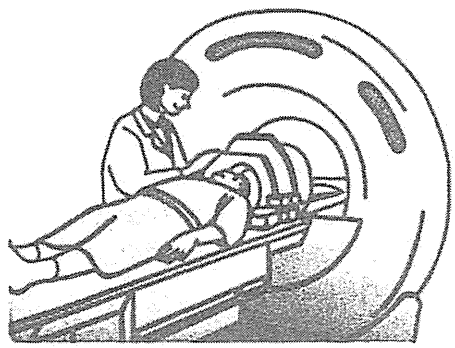
2．検診に10年間参加できそうな方（参加する意思があれば確証は不要）で、左下の「健康状態や病気に関する調査」に承諾していただける方のみ参加できます。

3．以下の方は参加できません。

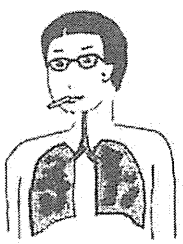
①いままで御自身が肺がんにかかったことがある人
②現在、肺がん疑いで医療機関で検査やフォローをしている人
③過去10年以内に「CTによる肺がん検診」を受診した人
④過去5年以内に、いずれかの「がん」にかかった人
⑤重篤な病気（重い心臓病、重い腎臓病など）にかかっている人

健康状態や病気に関する調査

この研究に参加される場合には、CTの方もX線の方も、後日あなたの健康状態や病気に関する調査を行わせていただく予定です。調査の方法は、ご本人あるいはご家族への手紙あるいは電話などによる問い合わせ、およびあなたが通院・入院される医療機関への調査ということになります。そのご承諾を得ることが、この研究への参加上必要ですのでご承諾をお願いします。

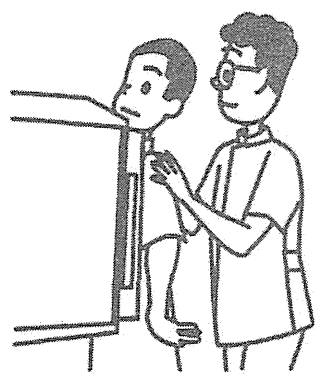


説明会および検診の日時：
平成22年　10月8日（金）
　　10月15日（金）
　　10月24日（日）
いずれも受付は9—16時
場所：羽咋市役所横体育館



腹部内臓脂肪CT検査について

最近メタボリック症候群が話題にのぼっています。X線検査の群の方は、この研究と並行して行う「採血＋腹部CTによる内臓脂肪と健康との関係を長期間観察する研究」に無料で参加することもできます。その研究では「採血」「生活習慣調査」「腹部CTによる内臓脂肪検査」を一度だけ行いますので、御希望の方は、お申し出ください。希望しない方は参加しなくて結構です。胸部CTの群の方は、予算の関係もあり両方のCTは受けられないので、その研究に参加することはできません。



参加御希望の方は、参加（仮）申込書に必要事項を記入して、参加（仮）申込書のみを返信用封筒に入れ、９月１８日必着でご返送ください。

厚生労働省垣添班　胸部CT検診小班（小班長：金沢医科大学教授　佐川元保）

事務局・問合せ先：金沢医科大学　呼吸器外科　佐川元保　電話＆FAX　076—286—1207
Email: sagawam@kanazawa-med.ac.jp

図1c.

参加（仮）申込書　（本申込書は、10月の説明会の時に書いていただきます）

研究名：低線量CTによる肺がん検診の精度および死亡減少効果評価のための個人単位ランダム化比較試験

参加するためには、説明書に載っている以下の記載に関して理解していただく必要があります。理解された場合には、右端の「理解した」の前の四角に☑を入れてください。

この研究はCT検診とX線検診とを比較するために行います	□ 理解した
研究は10年間の予定ですが継続できないこともあり得ます	□ 理解した
抽選でCTとX線に約半分ずつふりわけられます	□ 理解した
タバコを吸ってるかどうかで検査の内容がかわります	□ 理解した
CT検診では結果的に不要な検査や手術が行われる可能性があります	□ 理解した
精密検査や治療の際に合併症に巻き込まれる可能性はあります	□ 理解した
検診の費用は事務局負担、精密検査は保険診療で行われます	□ 理解した
私（参加者）の病気などの調査が行われることを承諾します	□ 理解した

事前調査（当てはまる方を丸で囲んでください。該当者は空欄を埋めてください）

現在通院中の医療機関がありますか？（ あり、なし ）
　ある場合には、その医療機関名（　　）病名（　　）
現在入院中または入所中ですか？（ はい、いいえ ）
　はい、の人は、その医療機関・施設名（　　）病名（　　）
過去10年間に入院したことがありますか？（ はい、いいえ ）
　はい、の人は、その医療機関・施設名（　　）病名（　　）
いままで御自身が肺がんにかかったことがありますか？（ はい、いいえ ）
現在肺がん疑いで医療機関で検査やフォロー中ですか？（ はい、いいえ ）
過去10年以内に「CTによる肺がん検診」を受けましたか？（ はい、いいえ ）
過去5年以内にどこかの「がん」にかかりましたか？（ はい、いいえ ）
現在重い病気（心臓病・透析中など）にかかっていますか？（ はい、いいえ ）

私は、上記を納得して、このプロジェクトに参加しますので、この書面で申込みます。

氏名＿＿＿＿＿＿＿＿＿＿＿＿　住所＿＿＿＿＿＿＿＿＿＿＿＿＿＿＿＿

第1希望(丸で囲む) 10/8(金)午前、10/8(金)午後、10/15(金)午前、10/15(金)午後、10/24(日)午前、10/24(日)午後

第2希望(丸で囲む) 10/8(金)午前、10/8(金)午後、10/15(金)午前、10/15(金)午後、10/24(日)午前、10/24(日)午後

第3希望(丸で囲む) 10/8(金)午前、10/8(金)午後、10/15(金)午前、10/15(金)午後、10/24(日)午前、10/24(日)午後

図2. 低線量CTによる肺がん検診の無作為化比較試験の仮参加申込書

30

3．説明会参加希望者に対して，3日間で9回（午前2回，午後1回）の説明会を行った。挨拶3分，ビデオ17分，口頭での説明25分，その後，登録という流れとした。説明会当日の配布書類は，①説明会の流れを説明したチラシ，②胸部CTによる肺がん検診の無作為化比較試験の説明書（郵送したものと同じ），③その研究の同意書および問診票，④腹部内臓脂肪コホート研究の説明書（こちらの研究の同意書と問診票は，X線群に振り分けられた後で内臓脂肪コホート研究参加の意思を確認してから配布），⑤検診受診者の意識調査および不安度調査，⑥説明内容の理解度調査，である。

4．必要書類（上記3の③と⑤）への記載を終えた順に，①記載内容チェック，②登録とランダマイズ，③検査の説明，④胸部CT群は撮影票記入後に胸部CT検査，内臓脂肪コホート希望者は問診票記入・身体計測・血圧測定・採血・腹部内臓脂肪CTの順にまわる，⑤高危険群は喀痰細胞診の容器を渡される，⑥必要書類を提出し，次年度の検査予定を書いたチラシを渡され帰宅，という流れで行った。

5．説明会には参加したが，不適格あるいは意思により研究へ不参加となった場合には，上記3の⑤⑥および，別に用意した「不参加理由書」で理由を選択または記載してもらってから帰宅，という流れとした。

研究への参加応諾率

329通郵送し，117通（35.6%）の返信があった。うち1例が重篤な心疾患で，別の1例がCTによる肺がん検診の10年以内の受診歴があり不適格となった。残りの115例に対し，説明会参加日時の割り振りを行い，その通知を郵送した。その結果，数名の変更希望があったほか，1名がどの日程でも参加不能なため研究参加を断念した。残りの114名（34.7%）が説明会参加予定となった。

説明会に参加した114名中，1名が仮参加申込以降にCTによる肺がん検診を受診したため不適格，1名が10年間の研究参加に難色を示し希望せず，1名がCT検診の不利益の可能性のため希望せず，計3名が不参加となり，残りの111名（郵送した対象全体の33.7%，返信された117例中の94.9%）が研究参加となった。

考　　察

このパイロットスタディの初年度の目的は，①研究全体の流れにおける多種の書類・ツールを作成する，②研究の実際における問題点を明らかにして計画を改善する，③対象者の何割が研究に参加するかを把握する，の3点であった。

必要な書類・ツールについては，研究の準備を進めて行きながら必要に応じて多種のチラシ，説明書，添え書き，データベースファイル，問診票などを作成した。今回のパイロットスタディは小規模ではあるが，大規模に進める場合にも充分対応できる基礎的な資料を作成し得たと思われる。同様に，種々の状況で発生した問題にその場で対応し，その後の研究計画の変更に生かすことができた。

研究参加応諾率は対象の1/3に達し，この種の検診の研究への応諾率としては，大変高かった。その理由としては，第1に「本年度の肺がん検診受診者」を対象としたため，健康意識が高いのみならず肺がん検診への興味が高い集団に対してリクルートを行ったことが挙げられる。第2には羽咋市が日頃からがん検診への意識が比較的高い自治体であったため，充分な協力が得られたことが影響していた可能性がある。第3に，かつて「がん検診の無作為化比較試験は日本になじまない」と言われていた時代があったが，今や多くの住民は無作為化に関して拒否感を持たないようになったことが考えられる。最後に，胸部X線群に「内臓脂肪研究へ参加可能」という条件を付けたことが影響した可能性がある。

説明会参加者の約95%が研究参加に至った。このことは，説明会参加者のほとんどが適格症例であり，かつ説明会で詳しい説明を聞いた後も参加の意思が変化していないという点で，今回使用した勧誘の手紙は「適格症例の絞り込み」「ランダマイズなども含めた研究計画の説明」の両面において有効に機能していると見ることができると思われる。

今後，CT所見の読影・精密検査・治療・追跡，と研究を進めていく予定であるが，CT検診での初回受診者の要精査率は通常5−10%程度，肺癌発見率は1%以下と考えると，今回のパイロットスタディでは要精査以降の症例数はきわめて少数になると考えられ，パイロットスタディでできることの限界もある。一方で，今回の研究により，約1/3という高い研究参加応諾率が得られたことは，他の地区でも同様なリクルート方法を取れば，相当高い応諾率が得られることが期待される。2010年11月に米国NCIのホームページでNational Lung Screening Trialの結果概要に関する速報がなされ（http://www.cancer.gov/newscenter/pressreleases/NLSTresultsRelease：2010年11月6日アクセス），胸部CT検診による死亡減少効果を認めたとの報告がなされたが詳細は未だ不明であり，また，非喫煙者に対する効果も不明である。その点で，わが国における胸部CT検診の効果に関する評価研究を今後実行していく必要性は全く変わっておらず，充分な予算措置が講じられることが待たれる。

この研究の事務作業に御援助いただいた山本和子氏，検診実務で御援助いただいた向瀬芳野氏，和田正美氏に深謝いたします。この研究は，がん研究開発費「がん検診の評価とあり方に関する研究」班（主任研究者：垣添忠生）の胸部CT検診小班（佐川元保（小班長），祖父江友孝，西井研治，江口研二，中山富雄，林　朝茂，小林　健，佐藤俊哉，佐藤雅美，細井　牧，濱島ちさと，斎藤博，鈴木隆一郎，三澤　潤，柿沼龍太郎，田中　良）で研究計画を作成し，同研究班の活動の一部として実施された。

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Recruitment for "A pilot study of randomized controlled trial to evaluate the efficacy of lung cancer screening by thoracic CT"

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Objective: To evaluate the efficacy of lung cancer screening by thoracic computed tomography (CT), a randomized controlled trial was planned in Japan. The randomized trial was designed as follows: 1) participants were randomly assigned into 2 groups, CT group and XP group; 2) XP group would receive 10 times of lung cancer screening by chest x-ray annually for 10 years; 3) smokers in CT group would receive 10 times of lung cancer screening by thoracic CT annually for 10 years; 4) non-smokers in CT group would receive 3 times of lung cancer screening by thoracic CT and 7 times of chest x-ray during 10 years. A pilot study was performed to evaluate the feasibility of the trial.

Methods: A letter for recruitment to participate in the above trial was mailed to the citizens in Hakui City, who were 50-64 years old and underwent regular lung cancer screening using chest x-ray this year. In the letter we explained that 1) the efficacy of lung cancer screening by thoracic CT had not been proved yet; 2) only half of the participants could undergo thoracic CT screening; 3) thoracic CT screening might cause unfavorable consequences like radiation exposure, false-positives or overdiagnosis.

Results: Of 329 persons who received the letter of recruitment, 117 replied. After meeting with us for detailed explanation, 111 persons participated in the above randomized trial.

Conclusion: The compliance of recruitment is high (approximately one third) and the above trial may be feasible.

Key Words: lung cancer screening, early detection, efficacy, thoracic CT screening

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Use of "AminoIndex Technology" for Cancer Screening

Naoyuki Okamoto

Amino acids play a central role in many biological activities. Several recent studies have reported that plasma amino acids can be used as biomarkers to assess disease risk or progression, or to select proper treatment. This review summarizes recent clinical research using a novel approach of multivariate analysis called "AminoIndex Technology", which is based on plasma amino acids profile for cancer screening. A multicenter study was conducted to explore and validate the application of "AminoIndex Technology" to cancer screening for gastric, lung, colorectal, prostate, and breast cancers. AminoIndex® Cancer Screening (AICS) scoring involves evaluating cancer based on amino acid concentrations using 5 types of AICS values. AICS enables simultaneous testing for multiple cancers regardless of cancer or tissue type. Furthermore, because AICS can detect stage II (stage B) or earlier cancers and can easily be performed on a plasma sample, it can be conducted in conjunction with comprehensive medical examinations or regular health check-ups. It is expected that "AminoIndex Technology" will be applied to cancer screening and various other areas of clinical utility.

Key Words : plasma amino, cancer screening, multivariate analysis, biomarker

Amino acids are usually considered protein subunits or nutrients. However, recent advances in the metabolomics of amino acids and high-throughput analytical techniques have shown that amino acids in the body (e.g. in the blood) can be used as biomarkers for evaluating disease risk or progression and for selecting proper treatment. For example, one study has reported that the risk of developing diabetes mellitus can be predicted from the metabolite profiles of a combination of 3–5 amino acids in the blood[1]. These new biomarkers, which are derived from combinations of amino acid concentrations, allow prediction of the risk of developing diabetes mellitus even when adjusted for conventional insulin resistance-related indices such as fasting insulin levels, homeostatic model assessment of risk of insulin resistance, and a 75-g oral glucose tolerance test. Amino acid metabolism indices differ from conventional biochemical indices and therefore may be regulated by a different paradigm. I anticipate that further research into amino acid metabolism will provide new information about disease risk and progression.

Advances in analytical techniques are among the most important reasons why novel findings concerning amino acid metabolism, such as the ability to predict the risk of developing diabetes, have been reported over the last several years. Plasma amino acid concentrations have conventionally been measured using an amino acid autoanalyzer that combines ion-exchange chromatography and a ninhydrin reaction. However, this method is so time-consuming that it has been used only for specific purposes, such as clinical studies with limited blood samples or in the diagnosis of inborn errors of metabolism. Progress

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in amino acid analysis techniques has led to using liquid chromatograph mass spectrometry (LC-MS) more frequently than the conventional method and analysis using LC-MS has enabled high-throughput measurement of plasma amino acids. Moreover, with the spread of this technology an enormous database of information is now available and it provides many clinically important insights into amino acid metabolism. As a result, many new amino acid metabolism findings have recently been reported.

In this review, I will first briefly discuss the role of plasma amino acids in the body and then summarize recent clinical research using novel plasma amino acid biomarkers for cancer screening.

Functions of amino acids in the body

Amino acids account for approximately 20% of an individual's body weight so a 50 kg person has approximately 10 kg of amino acids, most of which exist as proteins (**Fig. 1**). Free amino acids, known as the amino acid pool, are found in cells, intercellular components, plasma, and other biological components. Amino acids, which are digested and absorbed from foods and then enter the amino acid pool, from which they are used for protein synthesis, and are later returned to the amino acid pool as a result of protein degradation or excreted in the urine or feces. In this turnover process, the body replaces its protein composition through metabolism every 2–3 months.

The concentration of free amino acids in the plasma, which is part of the amino acid pool, is precisely regulated by a variety of control mechanisms and is maintained at a constant level in healthy individuals. Many studies have shown that various disease states, such as hepatic or renal failure, Alzheimer's disease, psychological disorders, and inflammatory bowel disease, may alter the plasma amino acid balance through aberrations in these regulatory mechanisms[2-6]. Also, as amino acid metabolism occurs actively in the muscle, the liver, the brain, the kidney, and the small intestine, changes in the amino acid metabolism balance in these organs are reflected as changes in the plasma amino acid concentrations.

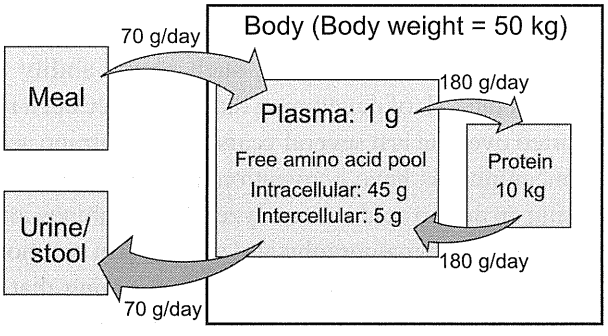


Fig. 1. Amino acid and plasma free amino acid pool in body

Metabolomics research and "AminoIndex Technology"

Recent advances in analytical techniques have enabled disease states to be analyzed through the comprehensive measurement of metabolites[1]. However, problems remain in the clinical setting with regard to the reproducibility, quantification, and cost of this type of analysis. The approach to such analysis using "AminoIndex Technology" is based on the assessment of diseases and physical conditions using plasma amino acid concentrations obtained through the measurement of a particular subset of metabolites. Measurement of amino acid metabolites is particularly useful for predicting various conditions because amino acid metabolism is closely related to many other metabolic pathways, such as glucose and lipid metabolism, and amino acids can therefore be considered as "hubs" to which many types of metabolites on the metabolic map are connected.

So the measurement of amino acid levels enables us to infer the statuses of various aspects of the overall metabolic map, such as those of glucose or lipid metabolism. In the previously mentioned study using metabolites as a measure of the risk of developing diabetes, an assay of 61 metabolites resulted in the identification of 3 or 5 metabolites associated with disease risk, all of which were amino acids. The underlying concept of "AminoIndex Technology" is to focus on these amino acids and to establish a reproducible and quantifiable method of measurement with applications in the clinical setting.

"AminoIndex Technology" is a technique in which multivariate analysis of the plasma amino acid concentration (**Fig. 2**) is used to compute disease or health condition scores. Through the use of statistical models, it can be used to predict the detection or severity of a single disease and clarify the status of multiple diseases or health conditions from a single blood sample. Recent research using "AminoIndex Technology" has been reported in other areas[7,8] in addition to the cancer studies reported in this review, and their clinical utility has been summarized in another review[9].

Application of "AminoIndex Technology" to cancer

Some authors have reported that plasma amino acid concentrations were altered in cancer patients because of various metabolic changes[10-14]. Furthermore, other clinical studies have demonstrated the possibility of using plasma amino acid concentrations as multivariate biomarkers in cancer screenings[15-18]. Furthermore, a particular clinical study on 5 types of cancer (gastric, lung, colorectal, prostate, and breast) was carried out to explore and validate the application of "AminoIndex Technology" to cancer screening[19].

The present multicenter study included the following institutions: Kanagawa Cancer Center; Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences; Osaka Medical Center for Cancer and Cardiovascular Diseases; Gunma Prefectural Cancer Center; Chiba Prefectural Cancer Center; Shizuoka Prefectural Cancer Center; Yokohama City University Medical Center; Yokohama Municipal Citizen's Hospital; Yokohama Minami Kyosai Hospital; Mitsui Memorial Hospital; Kameda Medical Center Makuhari and Kanagawa Health Service Association. It received institutional review board approval from all sites, and informed consent was obtained from all the patients. The present study used a new scoring system for calculation, known as AminoIndex® Cancer Screening (AICS), to analyze plasma samples from 2,043 cancer patients (**Fig. 3**) for screening purposes. Training and validation test datasets were used to establish an AICS score formula and to evaluate prediction accuracy, and all the results in this review are results in the validation test dataset. Amino acids included in the AICS formula for each cancer derived in the clinical research are shown in **Table 1**.

Some amino acids are commonly found in the AICS results for certain types of cancer: tryptophan (Trp) is seen in gastric, prostate, and breast cancers, whereas histidine (His) is seen in gastric, lung, and breast cancers. Similarly, particular amino acids may be cancer-specific, for example, threonine (Thr) in breast cancer or methionine (Met) in colorectal cancer. These data suggest that the AICS formula has plasma amino acid profiles that are common to several cancers or specific to a particular cancer.

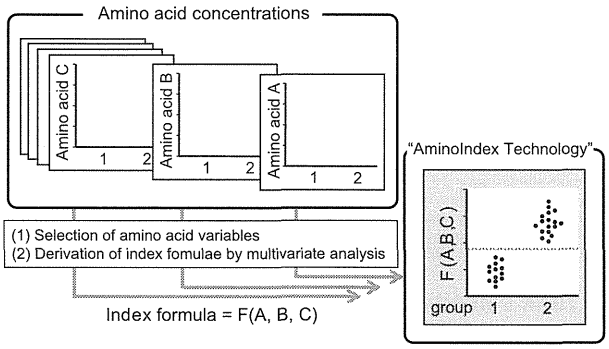


Fig. 2. Summary of "AminoIndex Technology"

AICS score and evaluation

AICS scoring involves evaluating multiple cancer types according to plasma amino acid concentrations on the basis of AICS values. As shown in **Fig. 4**, the minimum and maximum AICS values are 0.0 and 10.0, respectively, and the AICS values for specificities of 80% and 95% for each cancer are defined as 5.0 and 8.0, respectively. We presume that the higher the subject's AICS value, the greater the

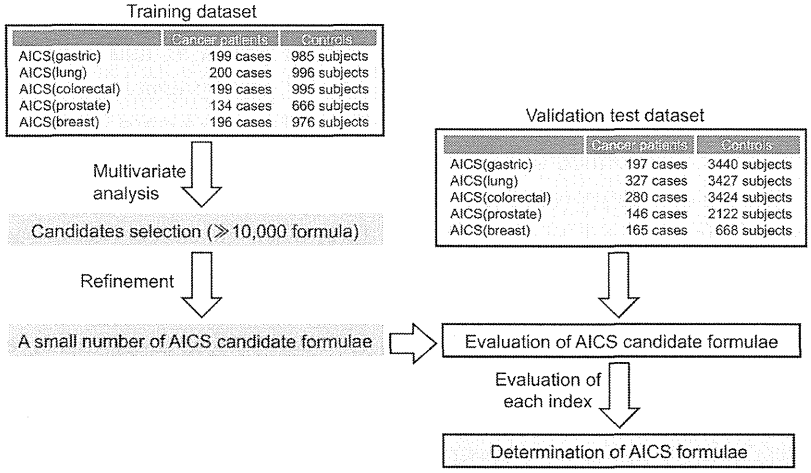
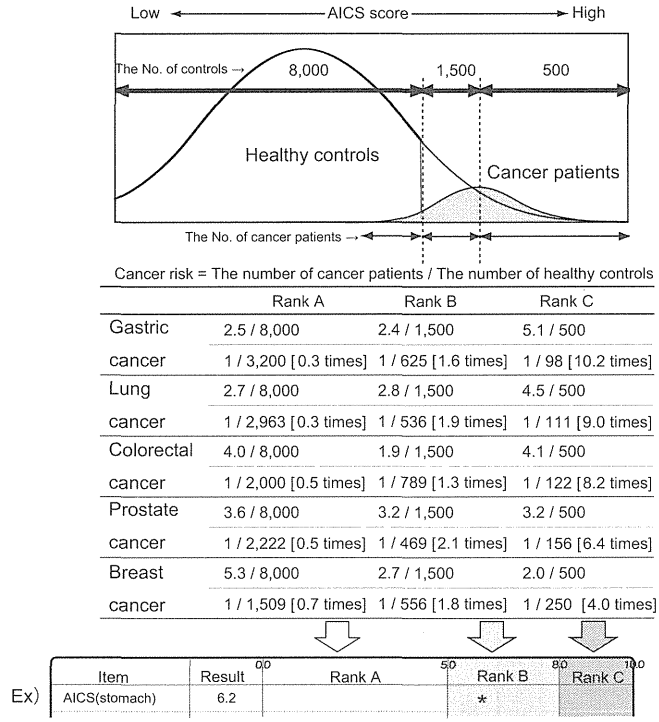


Fig. 3. Flow of AICS Derivation

Table 1. Amino acids included in AICS formulae

	Thr	Ser	Gln	Pro	Ala	Val	Met	Ile	Leu	His	Trp	Orn	Lys	Arg
AICS(gastric)					▼	▼			▼	▼	▼		▼	
AICS(lung)		▲	▼		■					▼		▲	■	
AICS(colorectal)		■		■		▼	▼	■					▼	
AICS(prostate)			▼		▲						▼	▲	▲	▼
AICS(breast)	▲				▲					▼	▼	▲		■

AICS: AminoIndex® Cancer Screening.
▲: Amino acids significantly increasing in cancer patients ($p < 0.05$)
▼: Amino acids significantly decreasing in cancer patients ($p < 0.05$)
■: Amino acids showing no significant difference in cancer patients



Cancer risk = The number of cancer patients / The number of healthy controls

	Rank A	Rank B	Rank C
Gastric	2.5 / 8,000	2.4 / 1,500	5.1 / 500
cancer	1 / 3,200 [0.3 times]	1 / 625 [1.6 times]	1 / 98 [10.2 times]
Lung	2.7 / 8,000	2.8 / 1,500	4.5 / 500
cancer	1 / 2,963 [0.3 times]	1 / 536 [1.9 times]	1 / 111 [9.0 times]
Colorectal	4.0 / 8,000	1.9 / 1,500	4.1 / 500
cancer	1 / 2,000 [0.5 times]	1 / 789 [1.3 times]	1 / 122 [8.2 times]
Prostate	3.6 / 8,000	3.2 / 1,500	3.2 / 500
cancer	1 / 2,222 [0.5 times]	1 / 469 [2.1 times]	1 / 156 [6.4 times]
Breast	5.3 / 8,000	2.7 / 1,500	2.0 / 500
cancer	1 / 1,509 [0.7 times]	1 / 556 [1.8 times]	1 / 250 [4.0 times]

Fig. 4. AICS value and rank classification

*The table shows the approximate percentage of incidence for each cancer by rank. The results for each cancer are indicated from 2 different perspectives on 2 levels. The upper level of each row is the cancer risk calculated as the proportion of 10,000 examinees, as mentioned previously. The lower level is the cancer risk when the numerator is 1. Thus, the values in parentheses are the mean risk ratios when the prevalence of cancer is 1 (approximately 1/1,000).

likelihood that the subject is suffering from cancer. AICS values are divided into 3 categories: rank A, <5.0; rank B, 5.0–8.0; and rank C, ≥8.0. The rank B or C cutoff and rank C cutoff are defined as 5.0 and 8.0, respectively. Thus, if the specificity is 95%, then 5% of the healthy controls are assessed as rank C (a false-positive rate of 5%), whereas if the specificity is 80%, then 20% of the healthy controls are assessed as rank B or C (a false-positive rate of 20%).

The prevalence of cancer is approximately 0.1%, that is, 10 of 10,000 people. Based on the present multicenter clinical research, in the case of gastric cancer, for every 10,000 people going for an AICS test, the rank A, B, and C groups have approximately 2.5, 2.4, and 5.1 cancer patients in them, respectively. Thus, the percentages of cancer patients in the rank A, B, and C groups are 0.03% (2.5/8,000), 0.16% (2.4/1,500), and 1.02% (5.1/500), respectively. Therefore, when the cancer risk in each group is compared with the whole population (i.e., a prevalence of 0.1), the rank A, B, and C groups have approximately 0.3-, 1.6-, and 10-fold cancer risks, respectively. However, it should be emphasized that if a person is evaluated as rank B or C, they do not necessarily have cancer. Similarly, if a person is evaluated as rank A, they are not necessarily free of cancer.

Table 2. Rank classification and specificity, sensitivity, and positive predictive value of AICS values for various cancers

AICS	Incidence rate	AICS value ≥ 5.0 (Rank B or C)			AICS value ≥ 8.0 (Rank C)		
		Specificity	Sensitivity	Positive predictive value	Specificity	Sensitivity	Positive predictive value
AICS(gastric)	0.0917	80	75	0.34	95	51	0.93
AICS(lung)	0.0657	80	73	0.24	95	45	0.59
AICS(colorectal)	0.0820	80	60	0.25	95	41	0.67
AICS(prostate)	0.0690	80	64	0.22	95	32	0.44
AICS(breast)	0.0775	80	47	0.18	95	20	0.31

All data are presented as percentages (%). To calculate the positive predictive value, the estimated incidence rate in the national predicted prevalence by age group, which was derived from 15 population-based cancer registries in the monitoring of cancer incidence in Japan (1975–2005)[20], was used instead of the prevalence rate.
AICS: AminoIndex® Cancer Screening

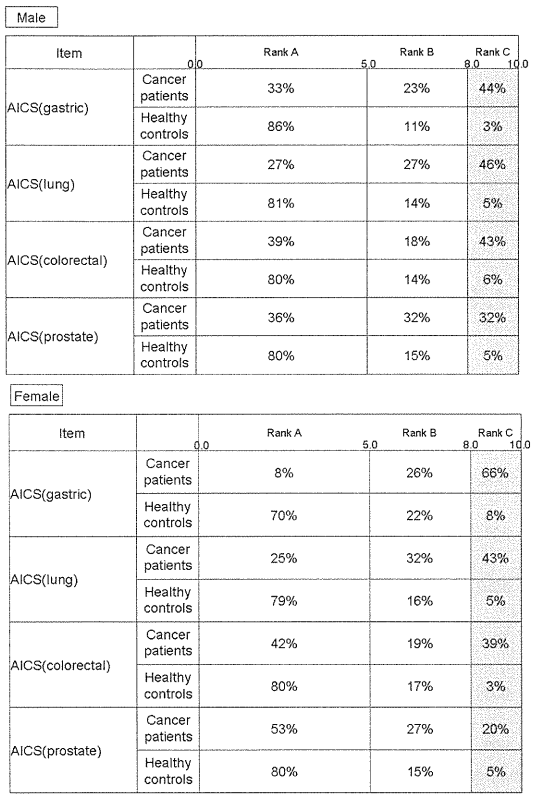


Fig. 5. AICS test result distribution

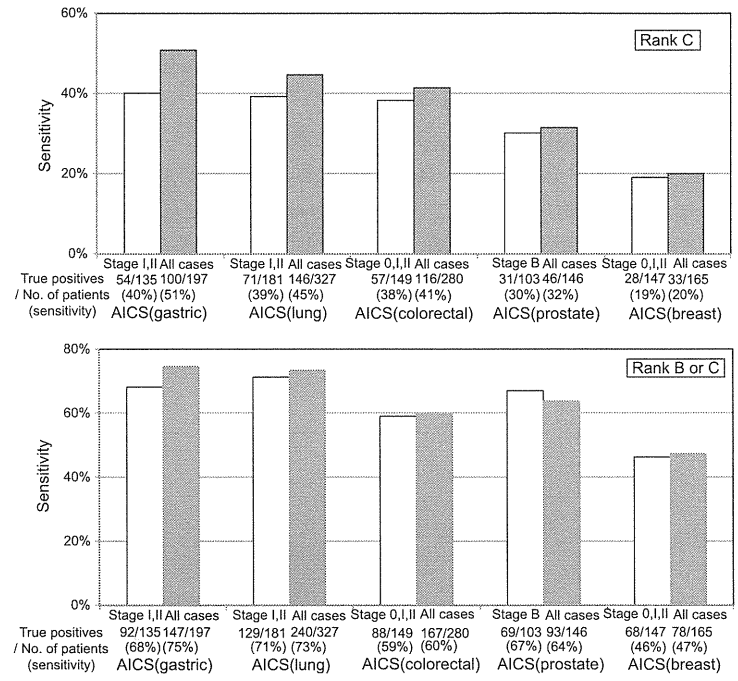


Fig. 6. Sensitivity of AICS for each type of cancer

Table 2 shows the AICS rank classifications and results (specificity, sensitivity, and positive predictive value). The sensitivities for gastric, lung, colorectal, prostate, and breast cancers at the rank B or C cutoff are 75%, 73%, 60%, 64%, and 47%, respectively, and those at the rank C cutoff are 51%, 45%, 41%, 32%, and 20%, respectively. **Fig. 5** compares the test results for the cancer patients and the healthy controls to demonstrate how the rank classification works, showing that 46% of the male patients with lung cancer and 5% of the male healthy controls were evaluated as rank C. The sensitivity for each cancer in patients with stage II (stage B) or earlier cancer is shown in **Fig. 6**. It shows that sensitivity for stage II (stage B) or earlier cancer is similar to that for cancer at any stage.

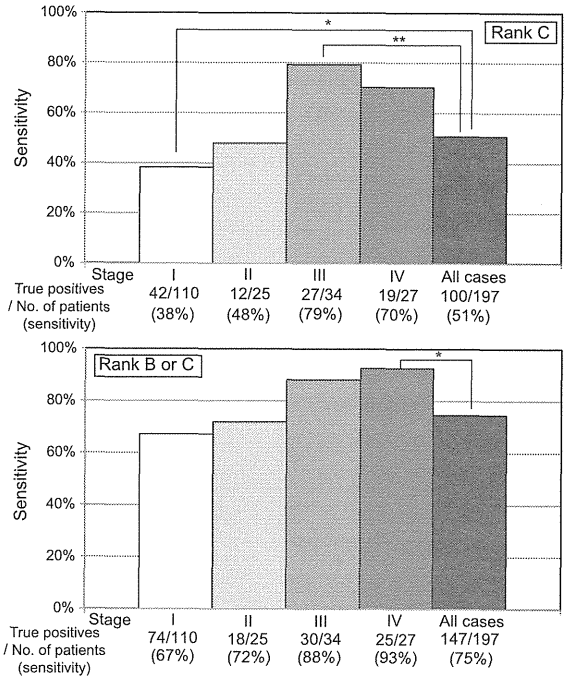
Evaluation of gastric cancer

Approximately 50,000 Japanese people died of gastric cancer in 2009, making it the second and third leading cause of death in men and women, respectively[20]. Although the pathogenesis of gastric cancer is still uncertain, *Helicobacter pylori* is thought to play a role. To use AICS for gastric cancer screening, an AICS

(gastric) score was derived from plasma amino acid levels in 199 gastric cancer patients using "AminoIndex Technology". Using this score, 197 patients in the validation test dataset, which was independent of the training dataset, were compared by either tumor stage, tissue type stratified analyses, or pepsinogen (PG) test results (**Fig. 7**).

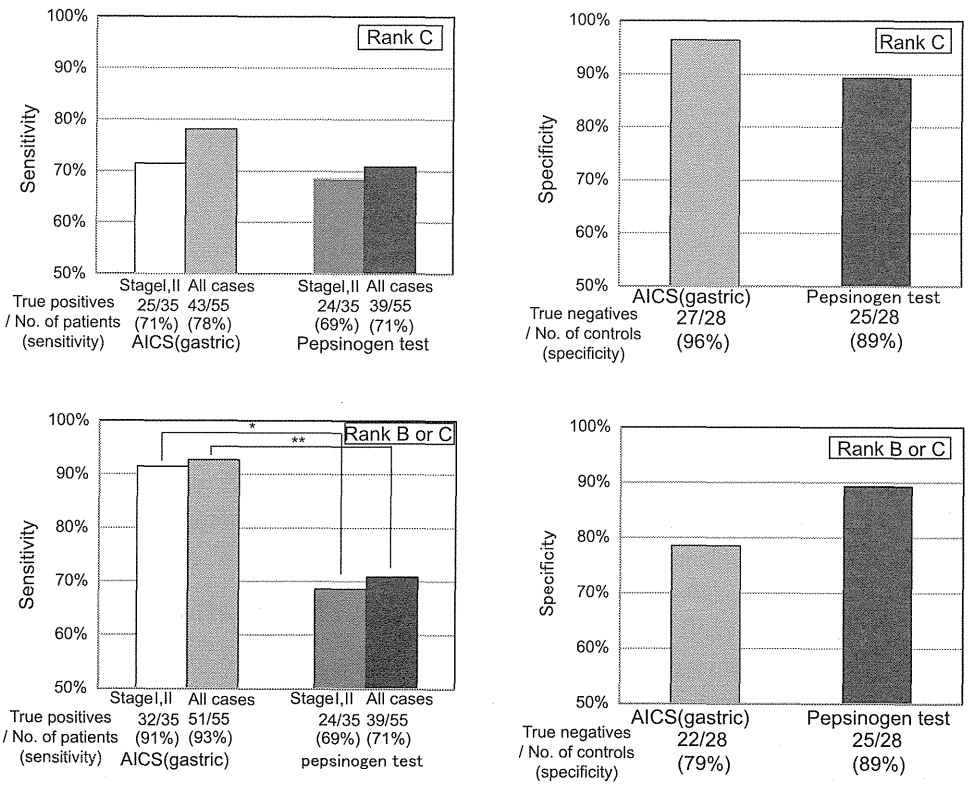
Early detection of gastric cancer is a major factor for a good prognosis. The sensitivity at each tumor stage is shown in **Fig. 7**. At the rank C cutoff, although there was a significant difference in sensitivity between stage I and all cases, the sensitivity was still 38% at stage I. There was no significant difference in sensitivity (48%) between stage II and all cases. At the rank B or C cutoff, there were no significant differences in sensitivity between stage I or II and all cases, and the sensitivities at stage I or II gastric cancer were high (stage I = 67%; stage II = 72%).

We compared the specificity and sensitivity of the PG test and AICS (gastric) in 55 patients with cancer and 28 healthy controls who underwent PG testing in a clinical study (**Fig. 8**). When PGI was ≤70 ng/mL and the PGI/II ratio was ≤3, the PG test result was defined as positive. When the sensitivities of PG testing and AICS (gastric) for gastric cancer were compared,



Fisher exact test: $^{*}p < 0.05$, $^{**}p < 0.01$

Fig. 7. Sensitivity of AICS (gastric) by stage



Fisher exact test: $^{*}p < 0.05$, $^{**}p < 0.01$

Fig. 8. Sensitivity and specificity for gastric cancer - AICS versus pepsinogen test

AICS (gastric) was significantly more sensitive ($p < 0.01$) than PG testing at the rank B or C cutoff.

As atrophic gastritis may produce a positive PG test, we compared the positive rate in atrophic gastritis patients using AICS (gastric) and PG testing (**Fig. 9**). In rank C patients, the positive rate for atrophic gastritis using the AICS (gastric) value was lower than that using PG testing ($p < 0.1$), suggesting that AICS (gastric) is a more accurate test for gastric cancer than the PG test.

We classified gastric cancer into several tissue types, which included poorly differentiated adenocarcinoma, signet-ring cell carcinoma, and tubular adenocarcinoma. Among them, poorly differentiated adenocarcinoma and signet-ring cell carcinoma are difficult to detect using PG testing. The results of differential analysis by tissue type are shown in **Fig. 10**. For AICS (gastric), there was no significant difference in sensitivity between tissue types at the rank B or C cutoff, although there was a significant difference in sensitivity among tissue types at the rank C cutoff. The lowest sensitivity for all cases was 43%. This was for tubular adenocarcinoma, which had the lowest sensitivity among the 3 tissue types. These data indicate that AICS (gastric) can be used as a screening method for at least 3 tissue types (poorly differentiated adenocarcinoma, signet-ring cell carcinoma, tubular adenocarcinoma).

Evaluation of lung cancer

Approximately 67,000 Japanese died of lung cancer in 2009, making it the leading and second leading cause of death in men and women, respectively[20]. Early detection of lung cancer is important because patient survival dramatically decreases as the disease progresses. To apply AICS to lung cancer screening, we used "AminoIndex Technology" to derive an AICS (lung) score from the plasma amino acid concentrations of 200 lung cancer patients. Using this score, 327 patients in the validation test dataset, which was independent of the training dataset, were stratified by stage or tissue type (**Fig. 11**). When AICS (lung) scores were stratified by stage (**Fig. 11**), we found no significant difference in sen-

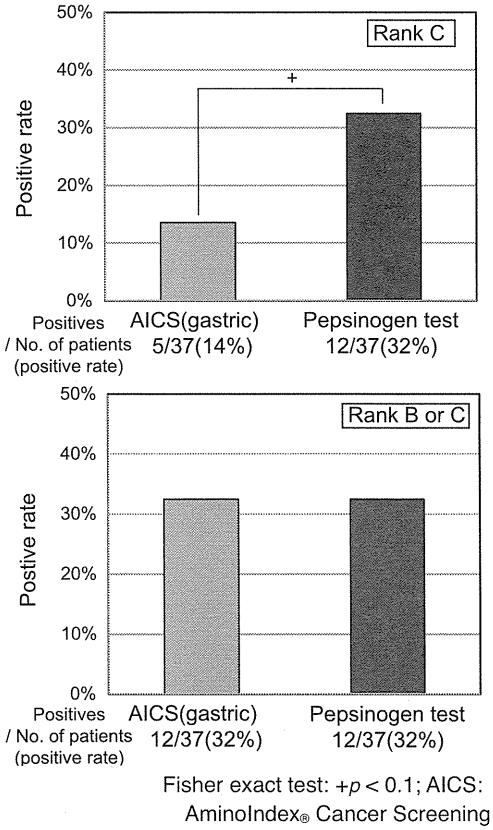


Fig. 9. Positive rate for atrophic gastritis

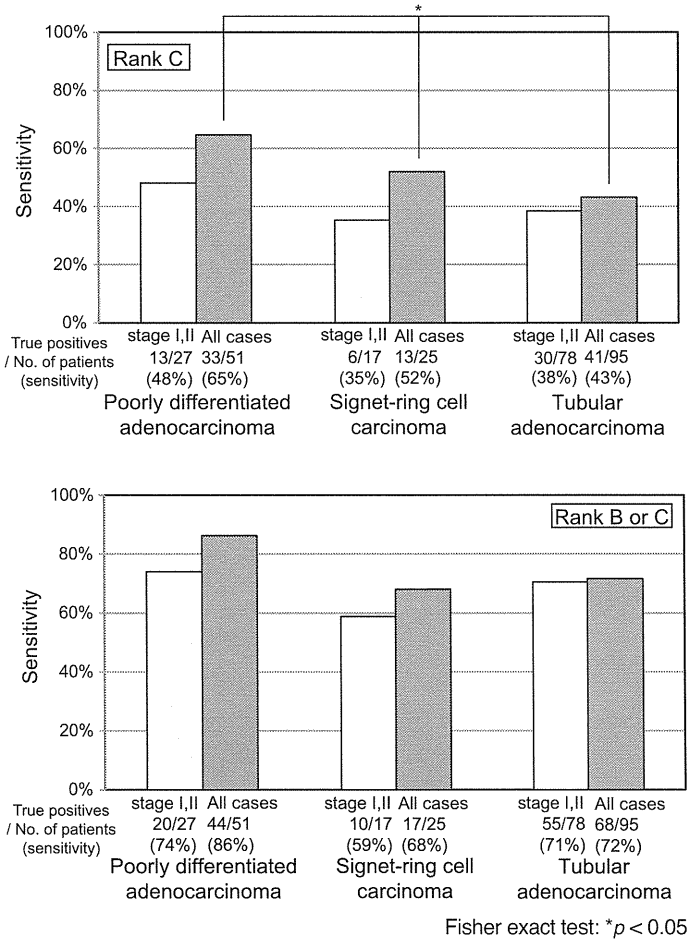


Fig. 10. Sensitivity of AICS (gastric) by tissue type

sitivity between stages. The sensitivity of stage I was 38% at the rank C cutoff and 70% at the rank B or C cutoff, suggesting that AICS (lung) can be used to detect early (stage I) lung cancer.

As lung cancer has a wide variety of tissue types and existing tumor markers are highly tissue-specific, it is difficult to determine tissue types other than squamous cell carcinoma using sputum cytology. To examine whether AICS (lung) scores were also dependent on tissue type, we stratified the AICS scores by tissue type (**Fig. 12**) and found no differences in sensitivities for adenocarcinoma, squamous cell carcinoma, or small cell carcinoma at the rank C cutoff and at the rank B or C cutoff. The sensitivities for all tissue types were more than 40% in rank C patients and more than 70% at the rank B or C cutoff.

Evaluation of colorectal cancer

Approximately 43,000 Japanese died of colorectal cancer in 2009, making it the third and leading cause of death in men and women, respectively[20]. Patient survival decreases as colorectal cancer progresses and therefore early detection of colorectal cancer is highly important. To apply AICS to colorectal cancer screening, "AminoIndex Technology" was used to derive an AICS (colorectal) score from the plasma amino acid concentrations in 199 patients with colorectal cancer. Using this score, 280 patients in the validation test dataset, which was independent of the training dataset, were stratified by stage and tissue type as shown below. When we stratified AICS (colorectal) sensitivity by stage (**Fig. 13**), we found no significant difference in sensitivity among stages. The sensitivity for stage 0 at the rank C cutoff was 55%, and that at the rank B or C cutoff was 64%. The positive rate of AICS (colorectal) for colonic polyps was significantly lower than that for colorectal cancer at the rank C cutoff and at the rank B or C cutoff (**Fig. 14**). This suggests that AICS (colorectal) is more specific for colorectal cancer than colonic polyps.

Evaluation of prostate cancer

Approximately 10,000 Japanese men died of prostate cancer in 2009[20]. Patient survival decreases as prostate cancer progresses and therefore early detection of prostate cancer is highly important. To apply AICS to prostate cancer screening, an AICS (prostate) score was derived from the plasma amino acid concentrations in 134 patients with prostate cancer using "AminoIndex Technology". Using this score,

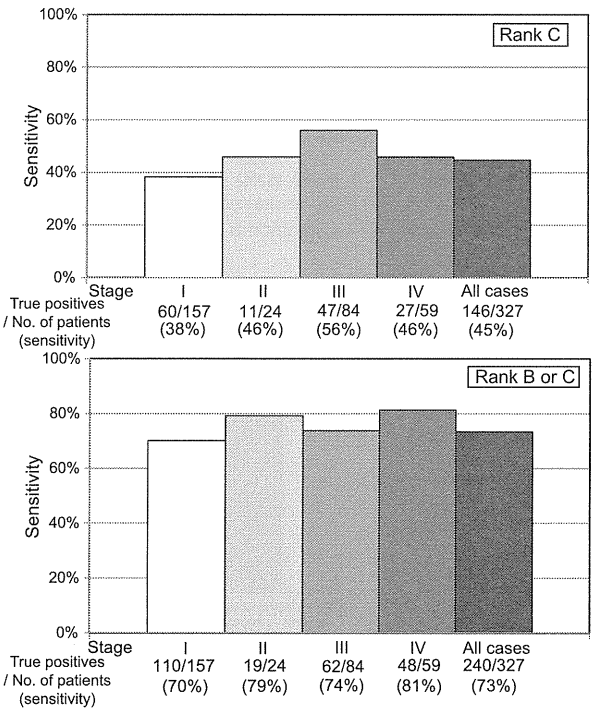


Fig. 11. Sensitivity of AICS (lung) by stage

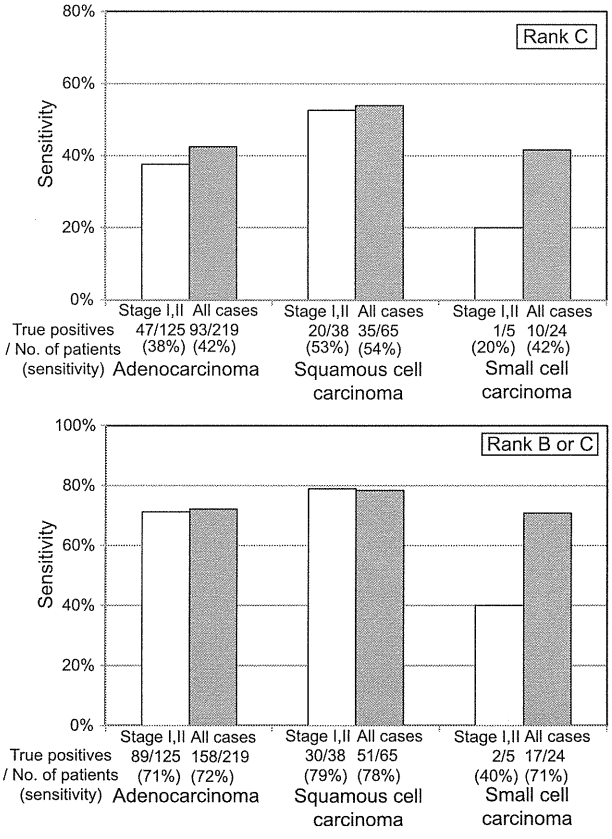


Fig. 12. Sensitivity of AICS (lung) by stage

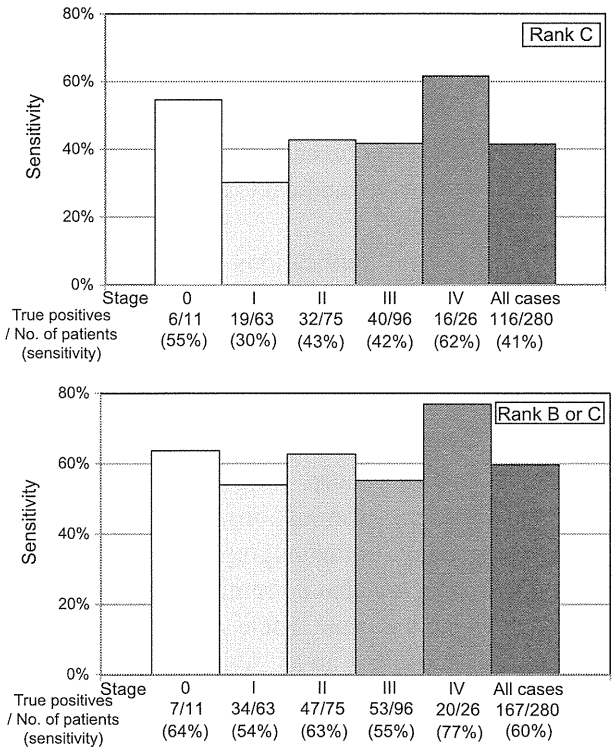
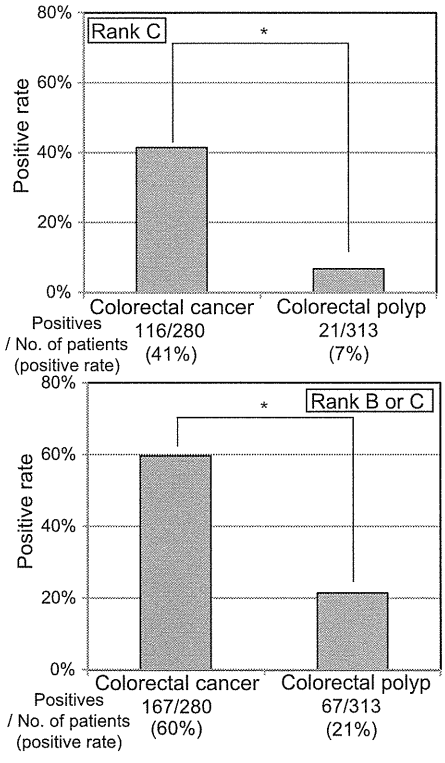


Fig. 13. Sensitivity of AICS (colorectal) by stage

146 patients in the validation test dataset, which was independent of the training dataset, were stratified by stage. The performance of AICS (prostate) in the "gray zone" of prostate-specific antigen (PSA) testing results was demonstrated.

When we stratified the AICS (prostate) scores by stage (**Fig. 15**), we found no significant difference in sensitivity among stages. The sensitivities at the rank C cutoff and at the rank B or C cutoff were 30% and 67%, respectively.

We also investigated a relationship between AICS (prostate) scores and PSA test values, which are commonly used for early detection of prostate cancer. The reference value for PSA in healthy individuals is ≤4.0 ng/mL, and further examination is needed when this value is exceeded. However, PSA scores of 4–10 ng/mL (the "gray zone") are not sufficiently predictive, making a better clinical test for such patients highly desirable. In patients with prostate cancer with PSA scores in the "gray zone," the sensitivity of AICS (prostate) was 35% at the rank C cutoff and 67% at the rank B or C cutoff (**Fig. 16**).



Fisher exact test: $^*p < 0.001$

Fig. 14. Positive rate for colorectal polyps

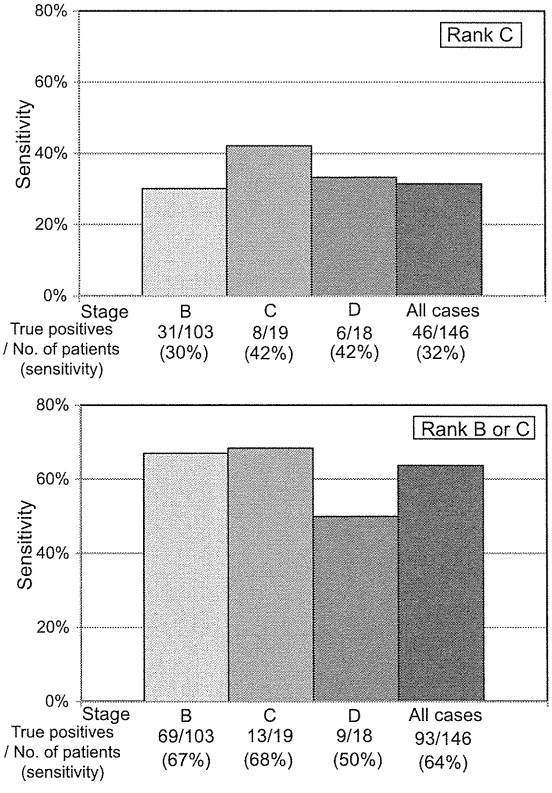


Fig. 15. Sensitivity of AICS (prostate) by stage

Evaluation of breast cancer

Approximately 12,000 Japanese died of breast cancer in 2009[20]. Early detection of breast cancer is very important, as the survival rate decreases with every stage. To apply AICS to breast cancer screening, an AICS (breast) score was derived from plasma amino acid concentrations in 196 patients with breast cancer using "AminoIndex Technology". Using this score, 165 patients in the validation test dataset, which was independent of the training dataset, were stratified by stage (**Fig. 17**). When stratified by stage (**Fig. 17**), the sensitivity of AICS (breast) was not significantly different between the stages. The sensitivity at stage 0 was 42% at the rank B or C cutoff.

Use of AICS

I would now like to summarize the results for each type of AICS test and describe its characteristics on the basis of the validation test dataset results (**Table 3**). AICS enables simultaneous testing for multiple cancers regardless of cancer or tissue type. Furthermore, because AICS can detect stage II (stage B) or earlier cancers and can easily be performed on a plasma sample, it can be carried out in conjunction with a comprehensive medical examination or regular health check-up.

There are several applications of AICS in clinical practice. First, it can be used as an alternative to existing cancer screening tests. Several well-known examination techniques are currently in use in cancer screening, for example mammography and ultrasonography in breast cancer screening. Depending on the type of cancer, various other screening tools, such as x-ray examinations, endoscopy, computed tomography, ultrasonography, and fecal occult blood testing are also currently used. The AICS method reviewed in this article can be applied to many cancer screening areas. AICS requires only a blood sample, making it more convenient and less invasive than several other screening methods. In addition to the screening methods mentioned above, genetic testing based on genetic polymorphisms is also used. However, one caveat regarding such genetic tests is that they cannot evaluate the contribu-

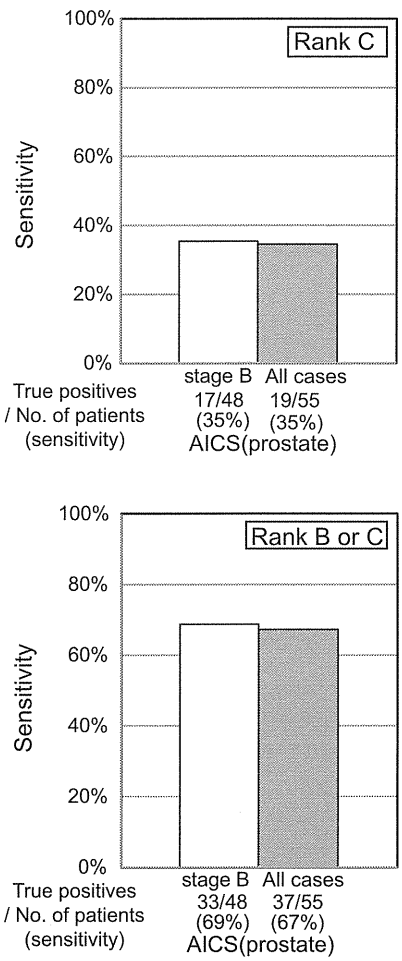


Fig. 16. Sensitivity of AICS (prostate) in PSA gray zone (4–10 ng/m)

PSA: prostate specific antigen; AICS: AminoIndex® Cancer Screening

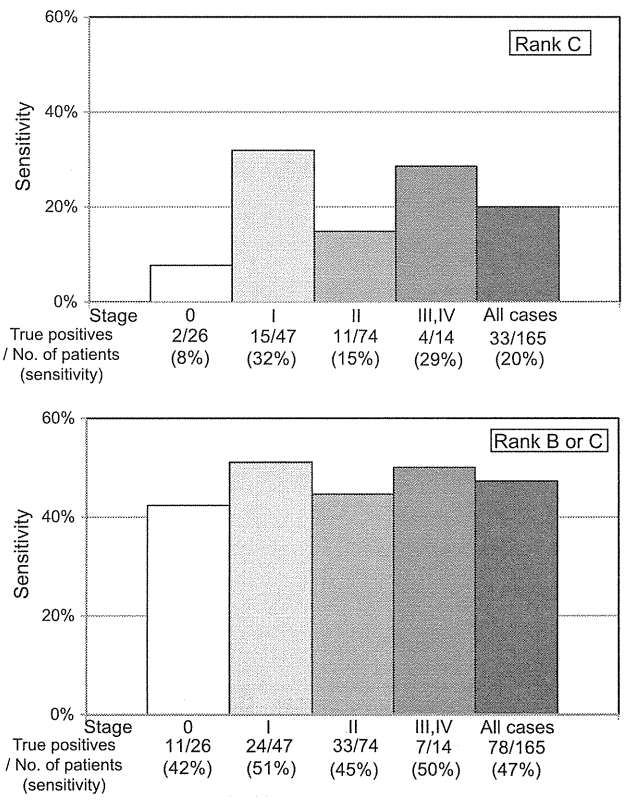


Fig. 17. Sensitivity of AICS (breast) by stage

Table 3. Characteristics of individual AICS tests

Test item	Characteristics
AICS(gastric)	1. High sensitivity for stage I and II gastric cancer 2. Higher sensitivity than pepsinogen testing in rank B or C 3. Lower positive rate for atrophic gastritis than pepsinogen testing in rank C 4. Equivalent sensitivity to tissue type, which is difficult to detect (poorly differentiated adenocarcinoma, signet-ring cell carcinoma)
AICS(lung)	1. High sensitivity for stage I and II lung cancer 2. Equivalent sensitivity for various tissue types of lung cancer
AICS(colorectal)	1. High sensitivity for stage 0, I, and II colorectal cancer 2. Low positive rate for colorectal polyps
AICS(prostate)	1. High sensitivity for stage B prostate cancer 2. High sensitivity for prostate cancer falling within the PSA gray zone (4–10 ng/mL)
AICS(breast)	1. High sensitivity for stage 0, I, and II breast cancer

AICS: AminoIndex® Cancer Screening; PSA: prostate-specific antigen

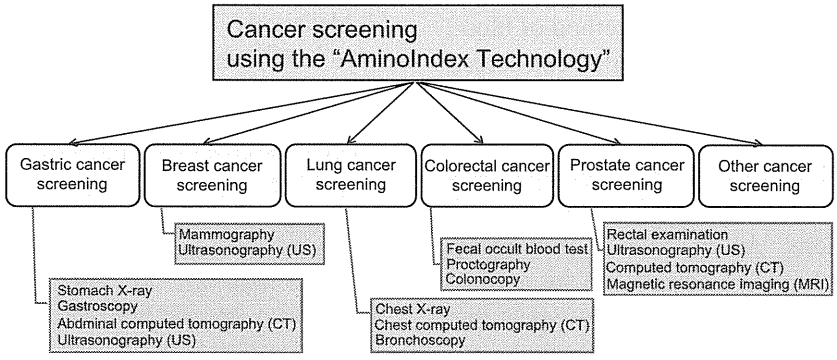


Fig. 18. Cancer screening using "AminoIndex Technology"

tion of environmental factors and lifestyle to overall risk. In contrast, as it is based on amino acid metabolites, AICS covers the influences of genetic and environmental factors and is therefore an alternative to genetic testing.

In addition, AICS can be used as a prescreening tool for specific cancers (**Fig. 18**). Existing screening tools have many drawbacks, such as exposure to radiation, cost, and inconvenience, reasons that can make people hesitant to undergo screening using them. With AICS, screening for gastric, lung, colorectal, prostate, and breast cancers can be conducted using a single blood sample, so AICS scores could be used to help a person decide whether to receive additional cancer screening.

In this article, we classified individuals whose AICS scores were ≥95% as rank C and those with AICS scores of ≥80% as rank B or C. However, it may be appropriate to use different classification thresholds according to the clinical context. After an AICS cutoff value is established based on appropriate specificity, it may be possible to apply it in practice to cancer screening.

Points to remember with AICS and issues to be addressed

This clinical research on AICS was conducted on Japanese subjects aged 25–90 years (for prostate cancer, 40–90 years). At present, it is not clear whether there are ethnic differences in AICS scores and therefore further studies are required. In addition, similar to regular medical examinations, blood must be collected in the morning after an 8 h fast because plasma amino acid levels are affected by dietary proteins and carbohydrates, as is the case of blood glucose and triglyceride levels. Not only solid food but also amino acid supplements (including liquids), amino acid preparations, and beverages containing protein and sugar (such as milk, soft drink and fruit juice) may influence results, if taken within 8 h before sampling. Also, as plasma amino acid concentrations differ during pregnancy, it may be

difficult to derive AICS values for pregnant women.

Conclusion

In this review, we have discussed the use of "AminoIndex Technology" in cancer screening. We expect that will be used as an alternative to current screening examinations or prescreening examinations for a variety of cancers. In the future, we hope that the effectiveness of AICS in practical cancer screening (e.g. cancer screening provided by local governments in Japan) will be clarified through clinical research, including longitudinal cohort studies.

Although we only discussed the application of "AminoIndex Technology" to cancer, clinical research on its use for other diseases is ongoing. If the results of such research verifies its usefulness, this technology will be established as a method of blood analysis using a single blood sample that can screen for multiple cancers while simultaneously evaluating the risk of developing many other diseases. In addition to evaluating disease risk, "AminoIndex Technology" could be used to promote dietary and exercise interventions. As mentioned previously, amino acid metabolomics research can be applied to many different clinical areas and we expect that novel applications for it will be created through such research.

Conflict of interest

I have no conflict of interest to declare for this review.

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Plasma Free Amino Acid Profiling of Five Types of Cancer Patients and Its Application for Early Detection

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Abstract

Background: Recently, rapid advances have been made in metabolomics-based, easy-to-use early cancer detection methods using blood samples. Among metabolites, profiling of plasma free amino acids (PFAAs) is a promising approach because PFAAs link all organ systems and have important roles in metabolism. Furthermore, PFAA profiles are known to be influenced by specific diseases, including cancers. Therefore, the purpose of the present study was to determine the characteristics of the PFAA profiles in cancer patients and the possibility of using this information for early detection.

Methods and Findings: Plasma samples were collected from approximately 200 patients from multiple institutes, each diagnosed with one of the following five types of cancer: lung, gastric, colorectal, breast, or prostate cancer. Patients were compared to gender- and age- matched controls also used in this study. The PFAA levels were measured using high-performance liquid chromatography (HPLC)–electrospray ionization (ESI)–mass spectrometry (MS). Univariate analysis revealed significant differences in the PFAA profiles between the controls and the patients with any of the five types of cancer listed above, even those with asymptomatic early-stage disease. Furthermore, multivariate analysis clearly discriminated the cancer patients from the controls in terms of the area under the receiver-operator characteristics curve (AUC of ROC >0.75 for each cancer), regardless of cancer stage. Because this study was designed as case-control study, further investigations, including model construction and validation using cohorts with larger sample sizes, are necessary to determine the usefulness of PFAA profiling.

Conclusions: These findings suggest that PFAA profiling has great potential for improving cancer screening and diagnosis and understanding disease pathogenesis. PFAA profiles can also be used to determine various disease diagnoses from a single blood sample, which involves a relatively simple plasma assay and imposes a lower physical burden on subjects when compared to existing screening methods.

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