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SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO

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Señor

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SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO
E. S. D.

Ref:	Expediente No.	**11-027.711**
	Patente de Invención:	**"COMPUESTOS DE TETRACICLINA SUSTITUIDOS CON FLÚOR EN C7"**
	Solicitud Internacional:	**PCT/US2009/053142**
	Clasificación Internacional:	**C07C 237/26, C07D 205/04**
	Titular:	**TETRAPHASE PHARMACEUTICALS, INC.**

RESPUESTA A REQUERIMIENTO OFICIO NO. 2443 NOTIFICADO POR FIJACION EN LISTA DE 4 DE FEBRERO DE 2014.

HUMBERTO RUBIO CAMACHO, mayor de edad, domiciliado en Bogotá, D.C., con la Oficina 506 de la Carrera 7 No. 74-64 de ésta ciudad, Abogado en Ejercicio, portador de la Tarjeta Profesional No. 50.007 expedida por el Consejo Superior de la Judicatura, actuando en carácter de apoderado de la sociedad **TETRAPHASE PHARMACEUTICALS, INC.,** domiciliada en 480 Arsenal Street, Suite 110, Watertown, Massachusetts 02472, Estados Unidos de America; reconocido por su despacho y encontrándome dentro de la debida oportunidad procesal, debidamente reconocido por su Despacho; encontrándome dentro del término legal para dar contestación al auto proferido por su Despacho mediante Oficio No. 2443, notificado por Estado de 4 de marzo de 2014, y cuya prórroga de término se radicó ante su Despacho el 30 de abril de 2014, me permito:

Manifestar:

I. En el informe oficial expedido contra la presente solicitud, se han emitido las objeciones que a continuación se resumen:

1. Determinación del Estado de la Técnica

El estado de la técnica relacionado con la presente invención se determinó con base en la fecha de presentación de la solicitud de prioridad, encontrándose los siguientes documentos, que según la Examinadora anticipan el objeto de esta solicitud:

Item	Documento	Título	Fecha de Publicación*
D1	EP0582789	Novel 7-(substituted)-8-(substituted)-9-(substituted glycyl) amido-6-demethyl-6-deoxytetracyclines	Febrero 16 de 1994
D3	EP0536515	Novel 7-substituted-9-substituted amino-6-demethyl-6-deoxytetracyclines	Abril 14 de 1993

El documento D1 divulga nuevos compuestos derivados 6 demetil, 6 deoxi tetraciclinas sustituidos 7,8 y 9 y sus composiciones, los cuales tienen efecto como antibióticos frente a microorganismos resistentes a las tetraciclinas.

El documento D3 divulga nuevos compuestos derivados 6 demetil, 6 deoxi tetraciclinas sustituidos 7 y 9 y sus composiciones, los cuales tienen efecto como antibióticos frente a microorganismos resistentes a las tetraciclinas.

2. Examen de Patentabilidad (Art 14 D486)

Nivel Inventivo (Art 18 D486)

Los documentos D1 y D3 se consideran el estado de la técnica cercano a la invención definida en la reivindicación 1.

El documento D1 divulga compuestos derivados 6 demetil, 6 deoxi tetraciclinas sustituidos 7,8 y 9 de fórmulas (I y II) y sus composiciones, con actividad antimicrobiana frente a un amplio espectro de microorganismos incluyendo los altamente resistentes a las tetraciclinas (pág. 1,2), en donde R3 y R4 son H y W es un grupo amino, o un heterociclo monocíclico (pirrolidina o azetidina, heterociclo bicíclico fusionado (azacicloalquil sustituido o no) con heteroátomos como el N.

La diferencia entre la invención, definida en la reivindicación 1, y el compuesto de D1 (fórmula I, pág 2) consiste en que no presenta sustitución en la posición 8.

No obstante la diferencia anterior, no es posible evidenciar un efecto técnico sorprendente. Por lo tanto, el problema técnico objetivo que pretende resolver esta invención se puede formular así: "cómo modificar el compuesto conocido en D1 para lograr una tetraciclina alternativa a las ya conocidas, con actividad antibacterial sobre bacterias resistentes a dichas tetraciclinas".

Sin embargo, D3 divulga el compuesto de formula (I) que no presenta sustitución en la posición 8 y divulga heterociclos en W (pirrolidina), asimismo la referencia (D2) divulga derivados de tetraciclinas sustituidos en C7 por un cloro (tabla 2, 7-cloroteraciclina), no están sustituidos en la posición 8, los cuales tienen actividad antibiótica sobre microorganismos resistentes a dichas tetraciclinas (Tabla 2, profilaxis terapia en humanos, pág.249).

Igualmente, es importante mencionar que en la descripción de la presente solicitud se evidencia la misma actividad farmacológica, como agente antimicrobiano, para el compuesto que se reivindica (tablas de las páginas 127 a 136) frente a los mismos microrganismos ensayados en D1 (tabla 1, pág.20 y 21) y D3 (tablas pág. 96 a 106).

En consecuencia, la persona del oficio normalmente versada en la materia, no sustituiría la posición 8 del compuesto de D1, de acuerdo a lo divulgado en los compuestos del documento D3 y a lo conocido también en la referencia D2 para llegar así al objeto de la reivindicación 1 de la solicitud. Por lo cual se considera obvia.

Puesto que las reivindicaciones dependientes 2 a 9 no mencionan características inventivas adicionales, tampoco se pueden considerar inventivas.

3. Conclusión

Los requisitos de Patentabilidad de la presente solicitud están afectados así:

Item	Documento	Reivindicaciones afectadas	Requisito que afecta
D1	EP0582789	1-9	Nivel inventivo
D3	EP0536515	1-9	Nivel inventivo

II.- En contestación a las observaciones anteriores, manifestamos lo siguiente:

- Reivindicaciones:

Acompañamos a esta respuesta un nuevo capítulo reivindicatorio (1-2), en el cual las reivindicaciones anteriores 1-7 se han eliminado, manteniéndose solamente las reivindicaciones 8 y 9, la reivindicación 8 anterior ahora la nueva reivindicación 1 dirigida al compuesto del Ejemplo 34 de la presente solicitud (comúnmente conocido como TP-434 o eravaciclina) y la reivindicación anterior 9 dirigida a una composición farmacéutica que comprende TP-434 y un vehículo o diluyente farmacéuticamente aceptable (ahora nueva reivindicación 2).

Debemos comentar que dichas reivindicaciones fueron concedidas mediante Resolución No.74812 de 30 de noviembre de 2012, y cuya novedad y nivel inventivo se encuentran en vigor desde la fecha señalada en la solicitud de patente tal como puede acreditarse en este mismo expediente.

La examinadora una vez realizo el estudio de patentabilidad, y ejecuto un análisis apropiado a la solicitud en estudio, encontró patentable las reivindicaciones 1 y 2 lo que la llevo a concederlas directamente sin emitir objeción alguna contra las mismas, por tanto solicitamos respetuosamente aplicar el mismo criterio y análisis que tuvo en esa oportunidad, pues la solicitud conserva su prioridad y en este corto lapso no se ha presentando un hecho que pueda afectar la patente que se caracteriza por tener en las reivindicaciones nivel inventivo y novedad, de lo contrario no se hubiera otorgado.

- Examen de Patentabilidad:

La Examinadora ha encontrado que los documentos de la técnica anterior D1 EP0582789 y D3 EP0536515 son relevantes para el nivel inventivo de la solicitud, por lo que concluye que su combinación hace obvio el contenido de la reivindicación 1 anterior.

La Examinadora argumenta que el compuesto actualmente reivindicado difiere de los compuestos divulgados en D1, el cual propone la solución más cercana a la presente

invención, en un sólo aspecto, la ausencia de un sustituyente en la posición 8 del sistema de anillos.

Por otra parte la Examinadora considera que la ausencia de sustituyentes en la posición 8, así como la sustitución de la posición 7 con un halógeno ya se divulgan en los documentos D2 y D3, de aquí que estas característica técnicas según la Examinadora que distinguen a la invención de D1, son evidentes por la combinación de las enseñanzas de estos documentos.

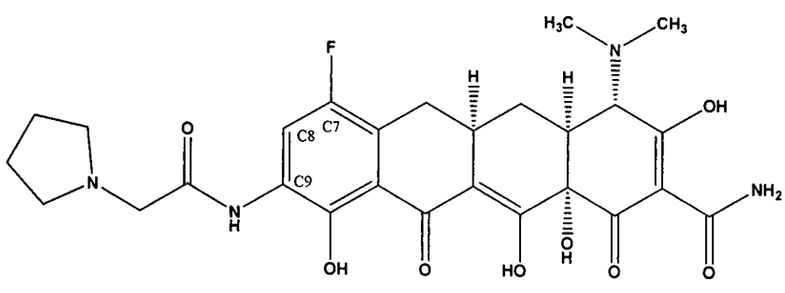
Sin embargo, una comparación de las estructuras divulgadas en los compuestos de D1 con la del compuesto de la nueva reivindicación 1 muestra otras diferencias adicionales fundamentales, las cuales como explicaremos posteriormente se relacionan con la presencia y la naturaleza de los sustituyente en la posición 7 y 9, sustituciones que no son obviamente derivadas en modo alguno de lo divulgado en los documentos D2 y D3 como asevera la Examinadora.

Notablemente ninguna de las referencias citadas por la Examinadora, ya sean consideradas por separado o en combinación, proporcionan la combinación específica de los sustituyentes se encuentran en las nuevas reivindicaciones 1 y 2, es decir, un sustituyente flúor en la posición C-7 y una 1-pirrolidinacetamida en la posición C-9.

Por otra parte, sobre la base de lo que se conocía sobre las tetraciclinas sustituidas con flúor en la posición C-7 (Anexo A) en el momento en que la presente solicitud se presentó, un experto en la materia no podría estar motivado a seleccionar el flúor como sustituyente en la posición C-7.

Adicionalmente, pruebas comparativas del compuesto de la reivindicación 1 (TP-434) y los del Ejemplo 5 de la patente US 5,574,026 (Backer et al.) se proporcionan en el presente documento, los cuales evidencian por la realización de ensayos comparativos que se presentaron durante la tramitación de la solicitud de patente análoga presentada para el territorio de los Estados Unidos, que los compuestos reivindicados (sustituidos con flúor en C-7 y con 1-pirrolidinacetamida en C-9) muestran una actividad inesperadamente superior sobre el compuesto derivado de tetraciclina estructuralmente similar de Backer *y otros* (sustituido con otro halógeno también en C-7, en este caso yodo, y con 1-pirrolidinacetamida en C-9).

El compuesto 34, objeto de las nuevas reivindicaciones 1 y 2, se reproduce a continuación:



(compuesto 34).

Seguidamente argumentaremos por qué el compuesto anterior es inventivo sobre las referencias citadas en el segundo informe del examen de patentabilidad de la solicitud, a saber los documentos D1 (EP 0582789) y D3 (EP 0536515).

- Con respecto a D1:

En primer lugar vale aclarar que en materia de fármacos, todo experto en la materia conoce bien que un pequeño cambio estructural en un compuesto puede tener gran repercusión en su actividad biológica, por lo que no solo la sustitución en sí, sino la naturaleza del sustituyente puede impactar considerablemente en la acción farmacológica que el mismo realiza.

El género químico definido por las reivindicaciones del documento D1, según la Examinadora el análogo más cercano a la invención, requiere y todos los compuestos ejemplificados en este documento así lo demuestran, la inclusión de un sustituyente en la posición 8 del núcleo de la tetraciclina. En contraste, compuesto reivindicado en la presente solicitud **no** incluye un sustituyente en esa posición.

Para establecer que las reivindicaciones 1 y 2, específicas para el compuesto 34 anterior, carecen de actividad inventiva, la Examinadora debe demostrar que una persona experta en la materia, sobre la base de las enseñanzas de D1, primero seleccionaría sustituyentes adecuados de las enseñanzas genéricas del mismo para la colocación en la estructura principal de la tetraciclina, y después modificaría estos sustituyentes para llegar a la invención reivindicada en la presente solicitud.

En particular, para llegar a la materia objeto de las nuevas reivindicaciones 1 y 2, una persona experta en la técnica tendría que, tras la lectura de D1, seleccionar el valor de R (sustituyente en C-7 de la fórmula I de D1) como flúor, seleccionar los valores de R3 y R4 en la fórmula I de D1 como hidrógenos, luego seleccionar el valor de W en la fórmula I como pirrolidin-1-il, y entonces eliminar el sustituyente obligatorio en C-8 (variable X de la fórmula I) y sustituirlo por un hidrógeno.

Discrepamos respetuosamente de la asunción de la Examinadora de que el documento D3 sugiere las modificaciones anteriores, pues de manera inadmisible la misma está haciendo uso de la retrospectiva para invalidar el nivel inventivo de la presente solicitud, ya que el documento D3 particularmente no sugiere el reemplazo en C-7 por flúor. Adicionalmente a esto, la combinación específica de un átomo de flúor en C-7 y de 1-pirrolidinacetamida en la posición C-9 tampoco se sugiere en D3, de aquí que estas dos características técnicas unido a la no sustitución en la posición C8 de la estructura principal, hacen notablemente diferente la invención con respecto a lo divulgado D1, y claramente las enseñanzas de D3 no conducen de manera obvia a estas tres posibilidades de cambio, sin que exista de por medio un esfuerzo inventivo adicional.

Es decir, considerando en detalles el contenido de D1, un experto en la materia no estaría motivado a seleccionar el flúor como un sustituyente en la posición C-7 ya que este documento divulga un sustituyente R genéricamente definido en C-7 y aunque este sustituyente R puede ser un halógeno, no hay ejemplificación de ningún tipo en

D1 de compuestos que tienen un grupo halo en la posición 7: los compuestos ejemplificados específicamente sólo poseen NMe_2 o H en C-7. De hecho, existe una clara preferencia en D1 de átomos de halógeno distintos de flúor. En efecto, véase, por ejemplo, la página 6, líneas 27-30 donde el flúor se omite de la lista de valores preferidos del sustituyente C-7.

Por lo tanto, si D1 se seleccionara por el experto en la técnica como un punto de partida para abordar el problema técnico objetivo de la presente solicitud, sin dudas no hubiera identificado aquellos compuestos que poseen un grupo 7-halo (por no hablar de un grupo 7-flúor) como los que ofrecen la mayor promesa para una solución. De hecho, incluso si una persona con experiencia en la técnica empezaría con uno cualquiera de los dos compuestos en donde W (parte del sustituyente C-9) es un C4 azacicloalquilo (es decir, los compuestos 43 y 62), no estaría motivado en sustituir el grupo H o $N(CH_3)_2$ presente en C-7 de estos compuestos con un flúor, en vista de lo anterior.

Es evidente que no hay nada en la divulgación de D1 que habría llevado a los expertos en la técnica a seleccionar específicamente un sustituyente en la posición 7 como flúor, porque D1 se centra en compuestos que tienen sustituyentes muy diferentes en esa posición.

- Con respecto a D3:

Las enseñanzas del documento D3 no resuelven las deficiencias de D1 por las siguientes razones:

Aunque el sustituyente X de D3 (véanse las fórmulas estructurales en la página 6 de D3) puede ser un halógeno, los únicos ejemplos en D3 que incluyen un sustituyente 7-halo son compuestos 7-yodo (Ver Ejemplos 20, 61, 66, 69, 74, 94). Las diferencias entre el flúor y el yodo (radio atómico, electronegatividad, etc.) son tales que un experto en la materia no esperaría que una molécula sustituida con flúor tuviera propiedades biológicas similares a las una molécula sustituida con yodo. Por otra parte, ninguno de los compuestos con sustituyente yodo en la posición 7 de D3 presenta además un resto pirrolidinacetamida en la posición 9, y en cada compuesto de D3 que tiene un heterociclo que contiene nitrógeno (por ejemplo, un pirrolidinacetamida) en la posición 9, el sustituyente en la posición 7 es un grupo amino, no es un halógeno. Por lo tanto, D3 no proporciona ninguna orientación para seleccionar flúor como sustituyente posición 7 y una pirrolidinacetamida como sustituyente en la posición 9 de forma combinada.

Por lo tanto, nada en D1 y D3, considerados juntos o en combinación, sugeriría a un experto en la materia a seleccionar el sustituyente 7-flúor junto con un sustituyente 9-pirrolidinacetamida.

Por lo tanto, si cualquiera de D1 o D3 fueran seleccionados por un experto en la técnica como punto de partida para abordar el problema técnico objetivo de la presente solicitud, el experto no hubiera identificado compuestos que poseen tanto un grupo 7-flúor como un grupo 9-pirrolidinacetamida como los que ofrecerían la mayor

promesa de una solución al problema técnico abordado, ya que ambas referencias se centran en compuestos que tienen muy diferentes combinaciones de sustituyentes en estas posiciones.

Con respecto al documento D2 (Chopra and Roberts, "Tetracycline Antibiotics: Mode of Action. Applications, Molecular Biology, and Epidemiology of Bacterial Resistance," Microbiology and Molecular Biology Reviews, June 2001, pp. 232-260 ("Chopra") citado en el primer reporte de examen de patentabilidad Chopra (D2) no va más allá de D1 en la dirección de una persona de habilidad ordinaria hacia el compuesto específico de las nuevas reivindicaciones 1 y 2. Los únicos compuestos en Chopra que incluyen tanto un sustituyente en C-7 y un sustituyente en C-9 son los compuestos que tienen $N(CH_3)_2$ en C-7 (Tabla 2). Aunque Chopra sugiere que la posición 8 (R_8 de la fórmula estructural reproducido en la figura 2 de Chopra) permanecerá sin sustituir, la misma figura también sugiere que la actividad antibacteriana óptima requiere que el sustituyente en la posición 7 (R_7 de la figura 2 de Chopra) esté ausente o se seleccione entre los grupos cloro, dialquilamino o nitro. Por lo tanto, no sólo Chopra no sugiere que el sustituyente en C-7 sea el átomo de flúor y C9 esté sustituido con 1-pirrolidinacetamida, sino que Chopra conduciría a un experto en la técnica lejos de un sustituyente flúor en C-7, ya que éste no se encuentra listado entre los "requerimientos de sustitución para la actividad antibacteriana óptima" (véase. Fig. 2).

En resumen, las enseñanzas de D1 están distantes de mostrar la posición C-8 no sustituida, y además este documento muestra una clara preferencia en excluir el sustituyente flúor en C-7, mientras que D3 y Chopra distan también de mostrar la colocación de un átomo de flúor en C-7, y por lo tanto de forma similar la combinación de un flúor en C-7 y una 1-pirrolidinacetamida en C-9. Ninguna de las referencias citadas durante el examen de patentabilidad de la presente solicitud, ni consideradas separadamente ni combinadas entre sí, conduciría a una persona con habilidad ordinaria en el campo técnico en cuestión a modificar cualquiera de los compuestos de los ejemplos descritos en las mismas de una manera necesaria para llegar al compuesto de las nuevas reivindicaciones 1 y 2. Por lo tanto dichas reivindicaciones son inventivas sobre los documentos citados.

Argumentos que refuerzan el nivel inventivo de la solicitud:

i. Una persona experta en el arte esperaría que el sustituyente 7-flúor reduciría la actividad antibacteriana de los compuestos de la invención.

Sobre la base en el conocimiento general disponible en el momento de presentación de la presente solicitud, una persona con habilidad normal en el campo técnico en cuestión esperaría que la modificación de un núcleo de tetraciclina mediante la colocación de un sustituyente flúor en la posición 7 resultara en la reducción de la actividad antibacteriana.

Como parte de esta respuesta presentamos el Anexo A, el cual es una publicación de Sato *y otros*. "Structure-Activity Relationship Investigation of Some New Tetracyclines

by Electronic Index Methodology", Los Alamos National Laboratory, Quantitative Biology, 1-18 (21 Aug. 2007).

Este anexo describe el uso de análisis computacional para predecir la actividad biológica de las tetraciclinas modificadas de diversa forma. Un conjunto de entrenamiento incluyó 14 derivados de tetraciclina conocidos con actividad antibacteriana reconocida (véase Tabla 1, página 2). El conjunto de entrenamiento se utiliza para generar un modelo teórico que se puede utilizar para predecir la actividad de los nuevos derivados de tetraciclina propuestos. Este modelo teórico produjo una puntuación que se clasificó cada derivado de tetraciclina propuesto como activas (A) o inactivo (I), basado en el gráfico a presentado en la Figura 5, página 9.

Los nuevos derivados de tetraciclina propuestas se describen a partir de la página 9, con referencias a las fórmulas estructurales genéricas que se muestran en la Figura 6, página 10, Figura 7, página 12, y la Figura 8, página 12. Para cada una de las tres fórmulas estructurales genéricas, treinta combinaciones de variables ("conjuntos estructurales") fueron investigadas, cada conjunto está definido por los valores de los sustituyentes indicados en la Tabla 4, página 10. Así, en total, noventa estructuras fueron investigadas (3 estructuras genéricas, con 30 combinaciones específicas de las variables para cada estructura genérica), y los resultados pueden verse en la Tabla 5, página 11.

En referencia a la Tabla 4, página 10, tres de las treinta combinaciones de sustituyentes en los "conjuntos de estructurales" incluyen grupos funcionales 7-flúor: 13, 14 y 20. De estas tres, el conjunto estructural 20 incluyó un átomo de flúor como el único valor que no es hidrógeno entre las cinco variables, permitiendo así aislar el efecto del flúor en la posición C-7 sobre la actividad biológica.

Haciendo referencia a la Tabla 5, basado en los "conjuntos estructurales" 13, 14, y 20, se consideraron nueve moléculas que incluyen el sustituyente 7-flúor: moléculas 13, 14, 20, 43, 44, 50, 73, 74, y 80. De estas, las moléculas 20, 50, y 80 incluyeron 7-flúor como el único sustituyente entre los cinco sustituyentes variables. Como la Examinadora podrá observar, las nueve moléculas que tienen la posición 7 sustituida con el radical flúor se prevé que son inactivas.

Por lo tanto, incluso si un experto en la materia seleccionara cualquiera de los compuestos descritos en D1 o D2 como un punto de partida para abordar el problema técnico objetivo de la presente solicitud, dicho experto habría esperado que la modificación de un núcleo de tetraciclina mediante la colocación de un sustituyente flúor en la posición 7 resultaría en la reducción de la actividad antibacteriana.

ii. Pruebas comparativas

A continuación se muestra una lista de documentos que se adjuntan a la presente respuesta y una breve descripción de cada uno de ellos.

Documento 1: Respuesta de 12.462.795 Primera Declaración-Agosto 2013.
Documento 2: 4142,1017-006 AAF presentado electrónicamente el 25/11/13.

Documento 3: Declaración Combinada 1 y 2 en el caso de método.

El Documento 1 incluye una Respuesta y Primera Declaración de la Dra. Joyce Sutcliffe, que fue presentada en el trámite de la solicitud análoga de los Estados Unidos, y en la cual se reivindica específicamente el compuesto TP-434 y una composición farmacéutica que comprende TP-434 (solicitud US 12/465,795). Los datos en el Anexo, se han generado para abordar un rechazo por obviedad de esta solicitud sobre el ejemplo 5 de Backer *y otros*. (Patente US 5,574,026) emitido por el examinador norteamericano. Esta presentación fue la primera presentación de datos comparativos. La revisión de los datos en el Anexo 5 muestra que en doce de las quince cepas bacterianas probadas, TP-434 fue de 2 a 8 veces más potente, y por lo menos 4 veces más potente para la mayoría de las cepas. Basándose en estos resultados, la Dr. Sutcliffe opinó que la potencia antibacteriana de TP-434 es inesperadamente superior en comparación con el compuesto divulgado por Backer.

El Documento 2 incluye una respuesta y Segunda Declaración de la Dra. Joyce Sutcliffe, también presentada en la solicitud de US que reivindica específicamente TP-434 y una composición farmacéutica que lo comprende. La Segunda Declaración fue presentada en noviembre de 2013 y se dirigió a las declaraciones hechas por la Examinadora en respuesta a la primera Declaración de la Dra. Sutcliffe. En la Segunda Declaración, la Dra. Sutcliffe se detuvo a esclarecer las razones científicas que apoyan la actividad inesperadamente superior del compuesto reivindicado sobre el ejemplo 5 de Backer *y otros*. En ella la Dra. Sutcliffe explica que las diferencias en los valores de CMI de TP-434 y el compuesto de Backer contra *E. coli* y *K. pneumoniae* son apreciables y demuestran una mejora sustancial, propiedad que es inesperada. Más específicamente, los valores de CIM del compuesto reivindicado (TP-434) contra *E. coli* y *K. pneumoniae* fueron suficientemente bajos (valores más bajos indican una mayor actividad) para continuar el desarrollo clínico del compuesto reivindicado como un antibiótico de amplio espectro, mientras que en el compuesto de Backer, los valores de CMI contra esas mismas cepas fueron demasiado elevados como para haber considerado el compuesto para su posterior desarrollo como un antibiótico de amplio espectro. Además, la Dra. Sutcliffe explica que la disminución entre 4 y 8 veces en el valor de CIM para el compuesto reivindicado en comparación con el compuesto de Backer se tradujo en la superioridad terapéutica relacionada con un aumento en la supresión de la resistencia a los antibióticos y la disminución de riesgo de toxicidad y otros efectos secundarios.

Debe considerarse además que en la página 2 de la respuesta presentada como parte del Documento 2, bajo el título "OBSERVACIONES" hay una discusión sobre la creciente amenaza para la salud pública de las infecciones resistentes a los antibióticos.

El Documento 3 incluye una respuesta presentada en marzo de 2014 en el caso de los métodos de tratamiento de infecciones bacterianas utilizando el compuesto TP-434 reivindicado. La respuesta se presentó a raíz de una entrevista personal con el examinador en la Oficina de Patentes de los Estados Unidos de América. Como parte de la respuesta, se presentaron la Primera Declaración y la Segunda Declaración de la Dra. Sutcliffe. Se acordó en la entrevista que este ensayo comparativo era

persuasivo y que la presentación de las Declaraciones resultaría en la concesión. Desde la presentación en marzo de esta documentación, el caso en US ya ha sido resuelto favorablemente para el solicitante.

Creemos, y la Dra. Sutcliffe así opina, que los datos presentados demuestran las propiedades inesperadamente superiores del compuesto reivindicado sobre los compuestos pertenecientes a la técnica anterior.

En consecuencia, podemos afirmar que existe doble razón para considerar el compuesto reivindicado como inventivo, pues su estructura no se deriva de la simple combinación de las enseñanzas descritas en los documentos de la técnica anterior citados como hemos demostrado, y en segundo lugar se ha evidenciado que el mismo presenta un efecto inesperadamente superior en comparación con compuestos estructuralmente cercanos en cuanto a su acción terapéutica relacionada con un aumento en la supresión de la resistencia a los antibióticos sobre un amplio rango de microorganismos patógenos

Anexos:

1. Capítulo reivindicatorio modificado (Reivindicaciones 1-2).
2. Anexo A: Sato F. *y otros*. Structure-Activity Relationship Investigation of Some New Tetracyclines by Electronic Index Methodology", Los Alamos National Laboratory, Quantitative Biology, 1-18 (21 Aug. 2007)
3. Documento 1: Respuesta de 12.462.795 Primera Declaración-Agosto 2013.
4. Documento 2: 4142,1017-006 AAF presentado electrónicamente el 25/11/13.
5. Documento 3: Declaración Combinada 1 y 2 en el caso de método.

Petición:

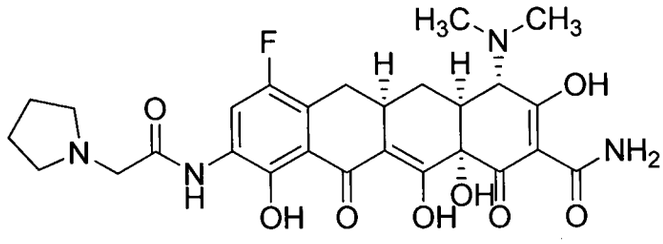
Conforme a lo anterior, una vez atendidos los requerimientos y exigencias de la Examinadora y al tener en cuenta el nuevo capítulo reivindicatorio presentado, solicito muy amablemente a su Despacho, que considerando los argumentos aquí expuestos, sea concedido el privilegio de invención solicitado, y en caso contrario se hagan las correspondientes observaciones con el ánimo de hacer lo pertinente dentro del presente trámite.

Atentamente,

HUMBERTO RUBIO CAMACHO
C.C. 17'192.062 de Bogotá
T.P. 50.007 Consejo Superior de la Judicatura
COD. 4134

REIVINDICACIONES

1. Un compuesto representado por la Fórmula Estructural (I):



(I)

o una sal farmacéuticamente aceptable del mismo.

2. Una composición farmacéutica, caracterizada porque comprende el compuesto de la reivindicación 1 o una sal farmacéuticamente aceptable del mismo y un portador o diluente farmacéuticamente aceptable.

Anexo A: Sato F. y otros.

XP-002548287

arXiv:0708.2931v1 [q-bio.BM] 21 Aug 2007

Structure-Activity Relationship Investigation of Some New Tetracyclines by Electronic Index Methodology

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February 1, 2008

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Abstract

Tetracyclines are an old class of molecules that constitute a broad-spectrum antibiotics. Since the first member of tetracycline family were isolated, the clinical importance of these compounds as therapeutic and prophylactic agents against a wide range of infections has stimulated efforts to define their mode of action as inhibitors of bacterial reproduction. We used three SAR methodologies for the analysis of biological activity of a set of 104 tetracycline compounds. Our calculation were carried out using the semi-empirical Austin Method One (AM1) and Parametric Method 3 (PM3). Electronic Indices Methodology (EIM), Principal Component Analysis (PCA) and Artificial Neural Networks (ANN) were applied to the classification of 14 old and 90 new proposed derivatives of tetracyclines. Our results make evident the importance of EIM descriptors in pattern recognition and also show that the EIM can be effectively used to predict the biological activity of Tetracyclines.

1 Introduction

Tetracyclines (TC) are a set of compounds used in human therapy against a broad-spectrum of microbial agents since 1947. They can be classified as low cost drugs with small side-effects being largely used by food industry and for therapy in animals and plants [1]. TC present activity against Gram-positive and Gram-negative anaerobic and aerobic bacteria, mycrobacterium and several protozoan parasites [2]. Non-antibacterial effects as anti-inflammatory, immunosuppressive [3], antioxidant [4] and anticancer [5, 6] are also attributed to them.

The excessive use of TC during years helped resistant bacteria proliferation, limiting then their clinical use. More than 30 TC resistance determinants have been described [7, 8] and several different TC resistance determinants have been identified in E. coli [9, 10, 11]. This emergence of resistant pathogens is becoming an increasingly important problem and it motivates the search for new TC variations and derivatives [12]. TC can be divided into two classes, the bacteriostatic typical TC and atypical TC, which are bactericidal and they exert their effects by promoting bacterial autolysis. Most probably due to their different modes of action, atypical TC have been shown to exert activity against some TC resistant bacteria [13, 14].

The chemistry of TC in solution is quite complicated [15] due to their ability to adopt different conformations, protonation states, and tautomeric forms, depending on the conditions investigated [16]. TC and its analogues undergo complex formation with a variety of the metal cations present in biological fluids [17, 18, 19, 20] and this affects their chemical as well as conformational equilibrium in solutions. Theoretical and experimental investigations

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C12

12

Figure 1: Basic structure of Tetracycline.

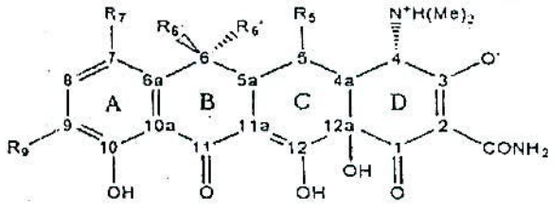


Table 1: Set of investigated Tetracyclines labelled accordingly to Figure 1 and respective biological activity index [27]. For compound 12 the substituintes on carbon 11 and 12 are interchanged and for compound 13 carbons 3 and 4 are linked to hydrogen atoms.

Molecule	Compound	R_5	R_6	$R_{6'}$	R_7	R_9	$K_{sens} \times 10^3$
01	Tetracycline	H	OH	Me	H	H	600.0
02	7-NH_2-6-Demethyl-6-deoxytetracycline	H	H	H	NH_2	H	190.0
03	5-Hydroxytetracycline	OH	OH	Me	H	H	650.0
04	9-NO_2-6-Demethyl-6-deoxytetracycline	H	H	H	H	NO_2	145.0
05	9-NH_2-6-Demethyl-6-deoxytetracycline	H	H	H	H	NH_2	350.0
06	7-Cl-6-Demethyltetracycline	H	OH	H	Cl	H	1100.0
07	7-Cl-Tetracycline	H	OH	Me	Cl	H	950.0
08	7-NO_2-6-Demethyl-6-deoxytetracycline	H	H	H	NO_2	H	1450.0
09	9-$N(CH_3)_2$-6-Demethyldeoxytetracycline	H	H	H	H	$N(Me)_2$	76.0
10	6-Demethyl-6-deoxytetracycline	H	H	H	H	H	250.0
11	6-Demethylamino-6-demethyldeoxytetracycline	H	H	H	$N(Me)_2$	H	250.0
12	5a,6-Anhydrotetracycline	H	Me	-	H	H	130.0
13	12a-Deoxytetracycline	H	OH	Me	H	H	8.5
14	7-Br-6-Demethyl-6-deoxytetracycline	H	H	H	Br	H	70.0

have contributed to the understanding of the interaction of TC with biological medium and their mode of action [21, 22, 23, 24, 25, 26]. Theoretical calculations have also been applied to investigations of new applications for old, new and hypothetical TC derivatives [22].

In this work we present an investigation of electronic and quantum features for a series of TC (Figure 1, Table 1) for which the biological activity against *E. Coli* is known and also to new compounds [27]. Our results showed that it is possible to directly correlate some quantum electronic descriptors to TC biological activity. This information is then used in the design of new compounds.

2 Methodology

We started our investigation with a detailed search for the global minimum of energy for 14 TC derivatives with known biological activity (Figure 1, Table 1). In the geometrical search of each molecule all degrees of freedom have been allowed to change during optimization and special atention has been devoted to the $NH(Me)_2$ group bounded to carbon 4, which is known to influence TC solution and gas phase stability [16]. All the calculations have been performed with semiempirical Austim Method 1 (AM1) [28] and Parametric Method 3 (PM3) [29, 30].

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The AM1 and PM3 methods are widely used in the investigation of organic molecules with very good results [31]. In this study one of the points we would like to evaluate is wheter the geometry and the correlation of theoretical properties with biological activity are dependent on the used semiempirical method. Molecules are studied in gas phase adopting the zwiterionic form, that is one of three TC basic forms and considered to be the active species in the biological medium. In zwiterionic form the total charge in molecule is equal zero but the groups bounded to the carbons 3 and 4 are charged and set to O^- and $NH^+(Me)_2$, respectively [27]. Although we can expect that Hartree-Fock methods such as AM1 and PM3 could have some difficulties in precisely discribing these forms, a systematic error is expected. Zwiterionic together with NH^- ionized species are commonly present in aqueous solutions and it is believed to complex with ions metals as Ca^{2+}, Mg^{2+}, Fe^{2+}, Zn^{2+}, Al^{3+} in biological medium, affecting the bioavailability and therapeutic TC properties [32, 33, 34].

For the geometry optimization and properties calculation we have used the Chem2Pac package [35]. Once optimized, the conformations of minimun energy were the basis for theoretical properties and electronic descriptors calculations. We based our structure-activity relationship (SAR) investigation on the Electronic Indices Methodology (EIM) and its descriptors. EIM was initially developed to identify the carcinogenic activity of polycyclic aromatic hydrocarbons [37, 36] using very simple rules. The methodology uses only two descriptors called Δ and η. The Δ is defined as the energy separation of molecular levels as indicated in Eq.(1).

$$\Delta = E_{level1} - E_{level2} \tag{1}$$

The descriptor η, is based on the Local Density of States (LDOS), which is obtained as the Density of States (DOS) for a selected group of atomic orbitals or atoms. The η is calculated as the LDOS difference between the most relevant molecular electronic levels (defined in the Δ calculation) over molecular regions identified as related to biological activity:

$$\eta = 2 \sum_{a=a_1}^{a=a_n} (|C_{a(level1)}|^2 - |C_{a(level2)}|^2) \tag{2}$$

C_a is the coeficient of atomic orbital from LCAO (Linear Combination of Atomic Orbitals) approximation. a_1 and a_n refer to atoms composing the selected molecular region. Δ contains global molecular information (involving the eigenvalues) while η contains local molecular information (LDOS) assumed to reflect the molecular environmental biochemical mechanisms (for details of EIM see refs. [37, 36, 38, 39, 35, 40, 41]).

With only these two descriptors EIM has classified with sucess, as active and inactive, molecules of differente classes of activity including steroid hormones [38, 39], integrase inhibitors [35], antibiotics [35], and tumoral [40] and antitumoral compounds [41]. Usually the EIM rules involve critical values assuming the boolean form: IF $\Delta > (<)\Delta_c$ AND(OR) $\eta > (<)\eta_c$, the molecule will be active, otherwise it will be inactive. Δ_c and η_c are critical values determined from EIM analysis. The critical values and EIM rules are automatically obtained from an exploratory scanning. In contrast to other SAR methods, EIM does not use training sets.

In order to obtain the EIM parameters and classificatory rules, the common molecular skeleton of the group of compounds investigated is divided into regions. Available informations about important molecular regions to determine biological activity is taken into account in the choice of the regions to the LDOS calculations. The present study we divided TC into 6 regions:

1. ring A and radicals;
2. ring B and radicals;
3. ring C and radicals;

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4. ring D and radicals, exception to groups $CONH_2$ and $N^+H(Me)_2$;

5. radical $N^+H(Me)_2$;

6. radical $CONH_2$.

The group of 14 molecules has been divided into two subgroups accordingly to their biological activity. Our criterion to classify molecules as active or inactive was based on the experimental values of K_{sens} index presented in Table 1. Molecules with K_{sens} lower than 100 are classified as inactive and those with K_{sens} greater than 100 as actives. The K_{sens} index represents the inhibition velocity constant based on cell growing velocity law [27].

The importance of electronic descriptors was evaluated by comparing EIM results with those obtained with standard SAR methods, Principal Component Analysis (PCA) and Artificial Neural Networks (ANN).

The PCA method is an extremelly useful explorative tool to separate samples into groups accordingly to their similarity and also to select important descriptors to these separations. In a geometrical interpretation each descriptor is considered a vector in a N-dimensional space and PCA method constructs a new space where new vectors, called principal components (PC), are linear combinations of the original descriptors. In the PCs space the samples are mapped through scores and the original descriptors through loading. From the loading plots, the important descriptors can be identified and the correlation pattern among them can be established. These calculations were carried out with the *Einsight* program [42]

The ANN analysis was carried out using the program package Perceptron-type Neural Network Simulator with back-propagation algorithm (PSDD) [43, 44]. PSDD consists of a three-layer perceptron network originally developed for drug design. This is a non-linear method and offers great advantage over conventional SAR methods for the cases where the target properties are not linearly dependent on the chosen parameters. In the ANN analysis we considered only the EIM descriptors to classify old and new proposed tetracyclines.

3 Results and Discussions

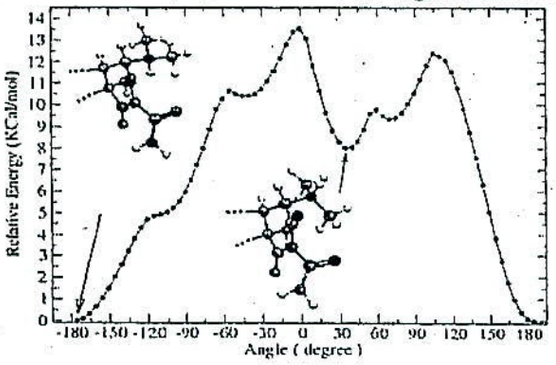
3.1 Structures

For the geometry optimization a conformational search was carried out considering the systematic variation of the dihedral angles related to the radicals bounded to the molecular skeleton. We initialy used the rotational step of 10 degrees to the OH, Me, NO_2, NH_2, $CONH_2$, and $NH(Me)_2$ groups in a rigid-rotor approach (one self consistent field calculation) using AM1 and PM3 hamiltonians. The ten best conformations (in terms of energy) were then selected and their geometries were fully optimized. All calculations (AM1 and PM3) have been performed with Mopac 6 implemented in Chem2Pac program [35] using high quality numerical precision optimization criteria.

An important characteristic of TC geometries is the presence of $NH^+(Me)_2$ group bounded to carbon 4. For the most stable conformations we investigated in details the energy variation related to the rotation of this group. For this specific part of study we optimized the geometry of each conformation generated by a rotation of 5 degrees at the dihedral formed by the hydrogen and nitrogen atoms of $NH^+(Me)_2$ group, carbon 4, and hydrogen bound on carbon 4 ($Dh_{NH^+(Me)_2}$). The complete rotation of 360° showed the existence of the $Dh_{NH^+(Me)_2}$ two local minima of energy. The structure at Dh=-180 degrees is named extended and the other at Dh=30 degrees twisted [24, 25, 16]. In Figure 2 we present the result obtained to molecule 01, for all the other molecules the curve of energy is similar. The relative energy is calculated considering the lowest energy as zero of reference. As can be observed from Figure 2 the lowest energy conformer corresponds to the dihedral $Dh_{NH^+(Me)_2}$ around 180 degrees. From the inset in the Figure 2 we can observe that in this conformation the hidrogen of $NH^+(Me)_2$ is directed toward the OH group at C12a. The second local minimum conformation is observed for $Dh_{NH^+(Me)_2}$ around 35 degrees and in this case the hydrogen atom points to O3 moiety.

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Figure 2: Relative energy for the rotation of dihedral $Dh_(NH^+(Me)_2)$ calculated with AM1 method to molecule 01. The -180 and 180 degrees correpond to the same conformation, the called extended conformation. The twisted conformation are located around 35 degrees. Structures in the inset show details of these conformations.

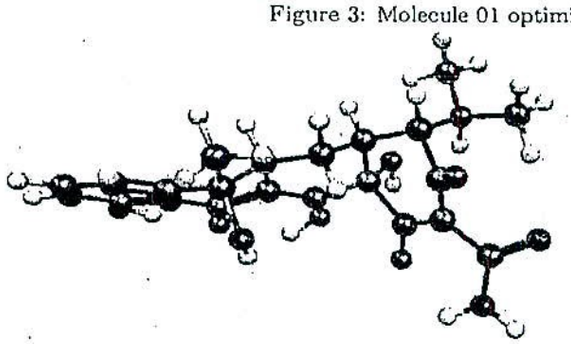


In order to better illustrate these conformations we show in Figure 3 the lowest energy conformer for molecule 01 in the extended conformation.

In Table 2 we present the main dihedral angles characterizing the TC rings from the global minimum of each compound calculated with AM1. Similar results were obtained with PM3 method.

As can be observed from the values of dihedrals 5-4a-12a-1 and 3-4-4a-12a (Table 2) all the molecules have the A ring non co-planar with the plane formed by rings D, C, and B. The rings C and B present a small torsion induced by the hydrogen atom bounded to carbon 5a. Torsions angle values between D-C and C-B rings are shown in second and third columns, respectively.

Figure 3: Molecule 01 optimized structure from AM1 calculation.



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Table 2: Main latex article templatedihedral angles (in degrees) for optimized TC structures with AM1 method. The second, third, and fourth columns show the planarities between rings D-C, C-B, and B-A, respectively. The fifth column indicates A ring angle torsions values. Labelling accordingly to Figure 1.

Molecule	7-6a-10a-11	6-5a-11a-12	5-4a-12a-1	3-4-4a-12a
01	-178.7	-138.5	-78.9	60.6
02	-175.7	-147.3	-84.8	60.1
03	-171.4	-121.6	-66.4	22.1
04	-178.1	-139.1	-78.6	62.4
05	-178.0	-139.7	-79.5	62.4
06	-179.7	-140.9	-79.9	60.5
07	-165.7	-141.7	-82.2	62.7
08	-174.5	-140.9	-80.1	60.7
09	-174.6	-140.0	-79.8	61.0
10	-178.9	-139.4	-79.2	62.4
11	-176.7	-139.8	-79.9	61.0
12	174.1	178.8	-72.3	59.8
13	-177.9	-136.7	-72.2	54.3
14	-177.8	-140.3	-79.9	60.9

3.2 The EIM Method and PCA Analysis

From the EIM analysis we obtained the Δ for pairs of frontier orbitals (Highest Occupied Molecular Orbital-HOMO, Lowest Unoccupied Molecular Orbital- LUMO and next closest levels) and the η data for all the regions listed in the methodology section. We observed that the best results for both AM1 and PM3 methods are obtained for ΔH (Eq. 3) and ηH (Eq. 4) calculated over the region 6 (radical $CONH_2$ bounded to carbon 2). In this case the equations 1 and 2 became:

$$\Delta = E_{HOMO} - E_{HOMO-1} \tag{3}$$

$$\eta = 2 \sum_{a=a_1}^{a=a_n} (|C_{a(HOMO)}|^2 - |C_{a(HOMO-1)}|^2) \tag{4}$$

The EIM parameter values are presented in Table 3. In order to build the EIM rules we determine the critical values of the descriptors ΔH_c and ηH_c. The classificatory rules are constructed trying to reproduce the experimental classes indicated in section 2 with relation to the K_{sens} values. For AM1 (Eq.5) and PM3 (Eq. 6) methods the following the rules have been obtained:

$$\text{IF } \Delta H \geq 0.70 \text{ AND } \eta H \geq 1.39 \text{ THEN molecule is inactive} \tag{5}$$

$$\text{IF } \Delta H \leq 0.58 \text{ OR } \eta H \geq -0.54 \text{ THEN molecule is active} \tag{6}$$

These crittical values (in equations 5 and 6) were obtained from an automatic screening search that determine the best statistical relevance of these numbers.

With these two descriptors and EIM simple rules for AM1 data it was possible to reproduce the experimental activity (active or inactive) with 100% of accurary. For PM3 calculation the rules achieved ~93% of accuracy, only molecule 11 was incorrectly classified.

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Table 3: EIM descriptors calculated from both AM1 and PM3 methods. Molecules are presented according to their K_{sens} values [27].

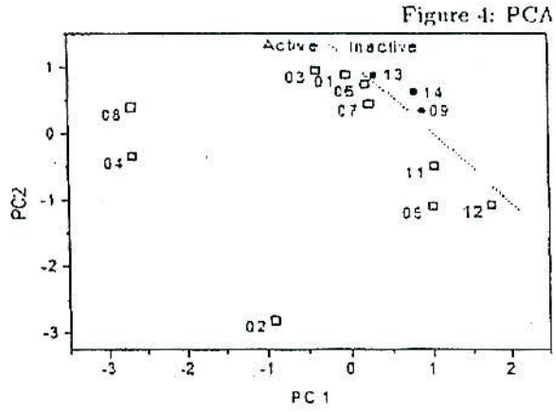
		AM1			PM3	
Molecule	$K_{sens} \times 10^3$	ΔH	ηH		ΔH	ηH
13	8.5	0.98	1.42		0.64	-0.56
14	70.0	0.89	1.39		0.93	-0.56
09	76.0	0.76	1.39	Inactive	0.59	-0.63
12	130.0	0.18	1.26		0.282	1.23
04	145.0	0.94	-0.59		0.465	-0.72
02	190.0	0.11	-1.38		0.194	-1.28
10	250.0	0.86	1.33		0.50	-0.71
11	250.0	0.39	1.39		0.599	-0.63
05	350.0	0.12	1.33		0.311	1.29
01	600.0	0.99	1.38		0.581	-0.63
03	650.0	1.00	1.38		0.893	-0.53
07	950.0	0.81	1.33	Active	0.575	-0.63
06	1100.0	0.94	1.34		0.465	-0.71
08	1450.0	1.18	-0.26		0.554	-0.63

The EIM results were then contrasted against the PCA methodology. For PCA analysis we considered a set of 25 parameters where electronic, physicochemical and stereochemical ones were included:

- ΔH
- ηH
- ΔL
- ηL
- Energy$_{HOMO}$
- Energy$_{HOMO-1}$
- Energy$_{LUMO}$
- Energy$_{LUMO+1}$
- LDOS (values from eigenstates HOMO, HOMO-1, LUMO, and LUMO+1)
- heat of formation
- surface area
- molecular volume
- hydratation energy
- Log P

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Figure 4: PCA - Scores from AM1 data.



- refractivity
- polarizability
- molecular mass
- electronic energy
- nuclear repulsion
- dipolar momentum
- ionization potential
- hardness.

Physicochemical variables frequently used in QSAR analysis have also been included in the calculations to evaluate the relevance of EIM descriptors. All the parameters were obtained from AM1 and PM3 calculations and PCA calculations have been carried out separatedly for them.

The data have been auto-scaled prior PCA analysis and we focused our attention on the two first principal components. The best results separating active and inactive compounds are presented in Figs. 4 and 5 to AM1 and PM3 descriptors, respectively.

Molecules in PCs planes are separated accordingly to the equations:

AM1

$$PC1 = -0.36(\Delta H) + 0.58(\eta H) + 0.74(LogP) \quad (7)$$

$$PC2 = +0.81(\Delta H) + 0.58(\eta H) - 0.06(LogP) \quad (8)$$

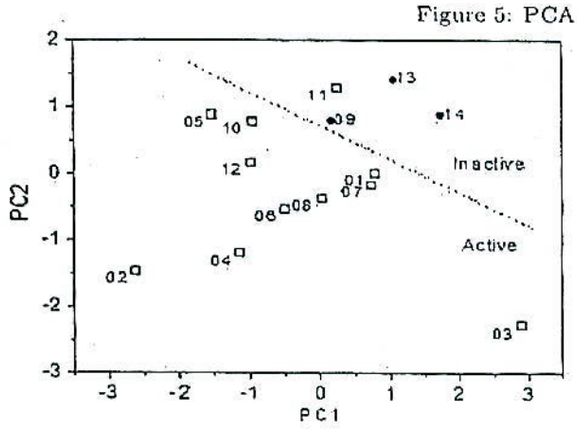
PM3

$$PC1 = +0.08(HOMO) + 0.66(\Delta H) - 0.42(HF) + 0.62(\eta H) \quad (9)$$

$$PC2 = +0.82(HOMO) + 0.04(\Delta H) + 0.54(HF) + 0.21(\eta H) \quad (10)$$

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Figure 5: PCA - Score from PM3 data.



For AM1 and PM3 the PC1 vs PC2 plane accumulates a total variance of 87.26% and 80.71%, respectively.

From Figures 4 and 5 and equations 7, 8, 9, and 10 we can see which variables are statistically more relevant among all the others in the set investigated. With exception of LogP all other variables are electronic and related to the EIM descriptors. Its is worth mentioning that during variables selection all variables were equally considered, no parameter scalling was used. In Fig. 4 a line delimiting the two classes of compounds was traced. For this case the molecules tend to agglomerate and the separation is not so clear. The results for PM3 data (Figure 5) show molecules more disperse and active and inactive groups separated. As already observed from EIM results, for the PM3 hamiltonian the molecule 11 was incorrectly classified as inactive.

These EIM and PCA results can be used together as an aplicable and fast tool in the classification of new Tetracyclines. In this sense we propose a set of 90 new hypothetical TC to what experimental data is unknown. We then used the EIM rules from the available experimental data to classify (active/inactive) these new hypotetical structures.

3.3 New Proposed Tetracyclines

We considered the structures of molecules 01, 12, and 13 as our basic skeletons (T_α, T_β, and T_γ) to generate new hypothetical TC (Fig. 6, 7, and 8). Combining these skeletons with common TC substituents (Table 1), plus some 7A column elements of the periodic table it was possible to generate 90 synthesis feasible new TC derivatives. Table 4 presents the 30 combinations of the substituents considered for each skeleton. The new 90 compounds where labelled as 01 to 30, 31 to 60, and 61 to 90 for substituents of Table 4 in T_α, T_β and T_γ, respectively.

These new compounds were then geometrically optimized following the same conformational searches described in section 2 and for the global minima we similarly obtained the EIM parameters values (ΔH and ηH) from group $CONH_2$ bound to carbon 2 (Table 5) accordingly to equations 1 and 2. For these new compounds we considered only AM1 data, since the EIM results proved to be very similar using AM1 or PM3 and level of accuracy for AM1 was 100%.

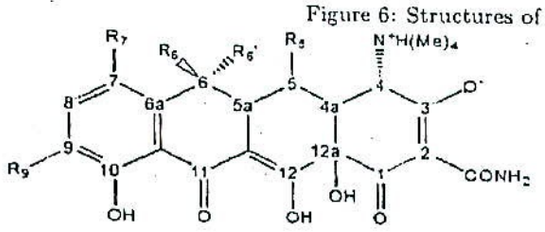
We use the EIM rules derived from AM1 calculations (equation 5) to classify the new TC as active or inactive (table 5). As can be observed from Table 5, 25 compounds are classified as actives while the remaining 66 are classified as inactives.

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Table 4: New proposed TC compounds. Labelling accordingly to Fig. 7 and 8.

Structural set	R_5	R_6	$R_{6'}$	R_7	R_9
01	Br	H	H	H	H
02	Br	H	H	H	Br
03	H	H	H	H	Br
04	H	H	Br	H	H
05	H	H	H	Cl	H
06	Cl	H	H	H	H
07	Cl	H	H	H	Cl
08	H	H	Cl	H	H
09	H	OH	H	Br	H
10	H	OH	Me	Br	H
11	H	OH	H	I	H
12	H	OH	Me	I	H
13	H	OH	H	F	H
14	H	OH	Me	F	H
15	I	H	H	H	H
16	I	H	H	H	I
17	H	H	H	H	I
18	H	H	H	I	H
19	H	H	I	H	H
20	H	H	H	F	H
21	H	OH	Me	NH_2	H
22	H	OH	Me	NO_2	H
23	H	OH	Me	H	NH_2
24	H	OH	Me	H	NO_2
25	OH	OH	OH	OH	OH
26	OH	H	H	H	$N(Me)_2$
27	H	OH	Me	H	$N(Me)_2$
28	H	H	H	OH	$N(Me)_2$
29	OH	H	H	H	NH_2
30	OH	H	H	H	NO_2

Figure 6: Structures of new proposed tetracyclines - T_α



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Table 5: EIM descriptors calculated to the new proposed TC and the proposed biological qualitative activity (QA). I and A refer to the inactive and active, repectively.

Molecule	ΔH	ηH	QA	Molecule	ΔH	ηH	QA	Molecule	ΔH	ηH	QA
01	0.82	1.39	I	31	0.80	1.39	I	61	0.84	1.42	I
02	0.80	1.39	I	32	0.90	1.39	I	62	0.83	1.42	I
03	0.86	1.39	I	33	0.93	1.39	I	63	0.87	1.42	I
04	0.93	1.39	I	34	0.87	1.39	I	64	0.95	1.42	I
05	0.83	1.39	I	35	0.84	1.39	I	65	0.84	1.42	I
06	0.82	1.39	I	36	0.81	1.39	I	66	0.84	1.42	I
07	0.76	1.39	I	37	0.89	1.39	I	67	0.78	1.42	I
08	0.96	1.39	I	38	0.89	1.39	I	68	0.97	1.42	I
09	1.13	1.23	A	39	1.04	1.39	I	69	1.00	1.42	I
10	0.93	1.39	I	40	0.91	1.39	I	70	0.94	1.42	I
11	1.04	1.38	A	41	1.03	1.39	I	71	1.05	1.41	I
12	0.97	1.39	I	42	0.94	1.39	I	72	0.98	1.42	I
13	0.91	1.39	I	43	0.93	1.39	I	73	0.91	1.42	I
14	0.84	1.39	I	44	0.84	1.39	I	74	0.86	1.42	I
15	0.82	1.39	I	45	0.79	1.39	I	75	0.84	1.42	I
16	0.86	1.39	I	46	0.90	1.39	I	76	0.88	1.42	I
17	0.91	1.39	I	47	0.93	1.39	I	77	0.92	1.42	I
18	0.94	1.39	I	48	0.92	1.39	I	78	0.95	1.42	I
19	0.90	1.39	I	49	0.85	1.39	I	79	0.92	1.42	I
20	0.78	1.39	I	50	0.81	1.39	I	80	0.79	1.42	I
21	0.05	1.39	A	51	0.16	1.39	A	81	0.08	1.42	A
22	1.18	-0.26	A	52	1.02	1.39	I	82	1.23	0.28	A
23	0.26	1.39	A	53	0.43	1.39	A	83	0.28	1.43	A
24	1.17	-0.50	A	54	1.19	-0.55	A	84	1.22	-0.44	A
25	0.68	1.39	I	55	0.83	1.39	I	85	0.73	1.43	I
26	0.85	1.39	I	56	0.86	1.39	I	86	0.85	1.42	I
27	0.03	1.39	A	57	0.19	1.40	A	87	0.25	1.43	A
28	0.13	-1.39	A	58	0.03	1.39	A	88	0.04	1.42	A
29	0.17	1.39	A	59	0.34	1.39	A	89	0.22	1.43	A
30	1.16	-0.38	A	60	1.17	1.33	A	90	1.22	-0.19	A

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Figure 7: Structures of new proposed tetracyclines - T_j

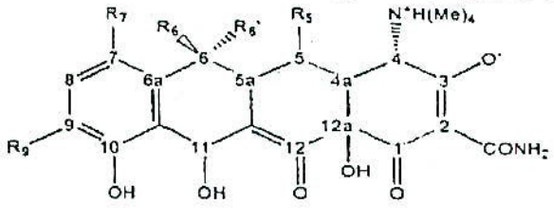
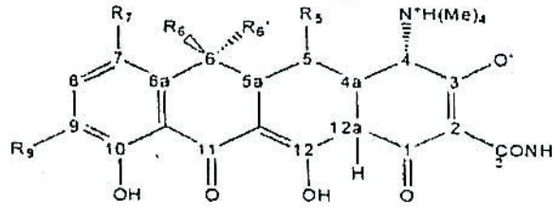


Figure 8: Structures of new proposed tetracyclines - T_7



The EIM and PCA methodologies consider the investigation of the correlation of theoretical descriptors with biological experimental data through linear combinations of variables. To verify the behavior of EIM descriptors (data from Table 3) in non linear correlations with experimental data we performed property-activity relationship searches using Artificial Neural Network methodology (ANN) [43, 44].

For ANN investigations we considered the 14 molecules studied in section III-A as the training group and the EIM parameters as network variables. In this trainning pattern we required the interactions precision of 10^{-5} and a 3×10^5 of interactions. In Table 6 we present the parameters used in ANN calculations, where α is a nonlinearity parameter of sigmoid functions of neurons in the second and third neuron's layer, θ is a threshold neuron parameter in a second and third layers, and ϵ is a constant weight mixing the weight matrix between recursive cicles.

In Table 7 we present the results for the training with the 14 molecules. The input data was set as class 1 to active molecules (1.000 0.000 - pattern) and class 2 to inactive molecules (0.000 1.000 - pattern). The columns four and five present the output values of training compared to the 0.000 and 1.000 values of input. We can observe that output values are very close to input, indicating a good training of the network. This good learning was achieved with precision parameters in cycle 242,477.

In Table 8 we present the results of the ANN classification, classes 1 and 2 correspond to active and inactive compounds, respectively. For comparision the EIM classification is also displayed. The results of EIM and ANN are

Table 6: Parameters of ANN for training pattern.

Layer	Neurons	α	θ	ϵ
1	3			
2	20	3.0	0.0	0.2
3	2	3.0	0.0	0.2

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Table 7: Input and output pattern of ANN training using EIM index of AM1 calculations of table 3.

No.	Trainning Pattern	Output Trainning	Class
01	1.000 0.000	0.996 0.004	1
02	1.000 0.000	1.000 0.000	1
03	1.000 0.000	1.000 0.000	1
04	1.000 0.000	1.000 0.000	1
05	1.000 0.000	1.000 0.000	1
06	1.000 0.000	1.000 0.000	1
07	1.000 0.000	0.997 0.003	1
08	1.000 0.000	1.000 0.000	1
10	1.000 0.000	1.000 0.001	1
11	1.000 0.000	1.000 0.000	1
12	1.000 0.000	1.000 0.000	1
09	0.000 1.000	0.000 1.000	2
13	0.000 1.000	0.005 0.995	2
14	0.000 1.000	0.003 0.997	2

in very good agreement. Only 6 compounds (11, 12, 39, 41, 52, and 71) out of ~90% (agreement) present conflictive classification. Interestingly all these 6 molecules are classified as active by ANN and inactive ones by EIM, which seem to have more restrictive conditions for active classification.

Finally, it is interesting to note the role played by substituents in the biological behavior of the new set of tetraciclines. According to our qualitative analysis the main substitution requirement for biological activity is the presence of NH_2 or NO_2 or dialkylamino groups at C7 and/or C9. Even for non-active derivative 13 (Table I) the presence of these substituents classifies as active new compounds 81-84 and 87-90 (Table VIII). Despite the whole molecular skeleton composed of 4 fused rings and the integrity of the ring A, the rest of the molecule including position C5 and C6 just plays minor role on the biological activity of tetracyclines.

4 Summary and Conclusions

In this work we applied the EIM methodology in the classification of activity of 14 derivatives of the Tetracicline and in the predictive classification of the activity of 90 hypothetical new derivatives. Our results showed that the EIM results are not method dependent indicating that the classification of activity of Tetraciclines is reproducible for different semiempirical methods. In comparison with conventional physicochemical descriptors the importance of EIM descriptors is reinforced by PCA and ANN methodologies. The rules constructed with EIM, PCA and ANN methods reproduced the experimental data with 100%, 96% and 100% of accuracy, respectivelly. For the new 90 derivatives proposed EIM and ANN are in good agreement with a percentage of 93% consistent classification. These results add to increasing data of EIM studies indicating the pure quantum electronic indices can be reliably used to classify the biological activity of large number of different classes of materials.

We hope the present study estimulate further experimental studies for tetracyclines in order to try to produce new antibiotic compounds.

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Table 8: Results of the ANN prediction to the new TC compounds using only EIM parameters. Classes 1 and 2 indicate active and inactives compounds respectively

	ANN			EIM		ANN			EIM		ANN			EIM
No.	Predict		Class	Class	No.	Predict		Class	Class	No.	Predict		Class	Class
1	0.002	0.998	2	2	31	0.001	0.999	2	2	61	0.000	1.000	2	2
2	0.001	0.998	2	2	32	0.017	0.983	2	2	62	0.000	1.000	2	2
3	0.001	0.999	2	2	33	0.015	0.986	2	2	63	0.000	1.000	2	2
4	0.044	0.959	2	2	34	0.001	0.999	2	2	64	0.000	1.000	2	2
5	0.001	0.999	2	2	35	0.000	0.999	2	2	65	0.000	1.000	2	2
6	0.002	0.998	2	2	36	0.001	0.999	2	2	66	0.000	1.000	2	2
7	0.001	0.999	2	2	37	0.007	0.993	2	2	67	0.000	1.000	2	2
8	0.383	0.633	2	2	38	0.002	0.998	2	2	68	0.000	1.000	2	2
9	1.000	0.000	1	1	39	1.000	0.000	1	2	69	0.087	0.911	2	2
10	0.066	0.938	2	2	40	0.005	0.995	2	2	70	0.000	1.000	2	2
11	1.000	0.000	1	1	41	1.000	0.000	1	2	71	1.000	0.000	1	2
12	0.924	0.080	1	2	42	0.032	0.970	2	2	72	0.002	0.998	2	2
13	0.016	0.984	2	2	43	0.021	0.980	2	2	73	0.000	1.000	2	2
14	0.001	0.999	2	2	44	0.001	0.999	2	2	74	0.000	1.000	2	2
15	0.002	0.998	2	2	45	0.001	0.999	2	2	75	0.000	1.000	2	2
16	0.005	0.995	2	2	46	0.015	0.985	2	2	76	0.000	1.000	2	2
17	0.007	0.993	2	2	47	0.015	0.986	2	2	77	0.000	1.000	2	2
18	0.138	0.870	2	2	48	0.004	0.996	2	2	78	0.000	1.000	2	2
19	0.006	0.994	2	2	49	0.001	0.999	2	2	79	0.000	1.000	2	2
20	0.000	1.000	2	2	50	0.000	1.000	2	2	80	0.000	1.000	2	2
21	1.000	0.000	1	1	51	1.000	0.000	1	1	81	1.000	0.000	1	1
22	1.000	0.000	1	1	52	1.000	0.000	1	2	82	1.000	0.000	1	1
23	1.000	0.000	1	1	53	1.000	0.000	1	1	83	1.000	0.000	1	1
24	1.000	0.000	1	1	54	1.000	0.000	1	1	84	1.000	0.000	1	1
25	0.004	0.995	2	2	55	0.001	0.999	2	2	85	0.000	1.000	2	2
26	0.002	0.998	2	2	56	0.001	0.999	2	2	86	0.000	1.000	2	2
27	1.000	0.000	1	1	57	1.000	0.000	1	1	87	1.000	0.000	1	1
28	1.000	0.000	1	1	58	1.000	0.000	1	1	88	1.000	0.000	1	1
29	1.000	0.000	1	1	59	1.000	0.000	1	1	89	1.000	0.000	1	1
30	1.000	0.000	1	1	60	1.000	0.000	1	1	90	1.000	0.000	1	1

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BNSDOCID: <XP_____2548287A_I_>

SMA/saj
August 23, 2013

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	12/462,795	Group:	1628
Filed:	August 7, 2009	Examiner:	Badio, Barbara P.
Confirmation No:	5619		
For:	C7-Fluoro Substituted Tetracycline Compounds		

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Commissioner:

This Reply and Request for Continued Examination-Prioritized Examination under 1.102(e)(2) is being filed in lieu of a filing a Brief on Appeal, pursuant to the Notice of Appeal received in the U.S. Patent and Trademark Office on January 23, 2013 in the above-identified application. An extension of time to file the Brief on Appeal is respectfully requested. A Petition for Extension of Time and the appropriate fee are being filed concurrently.

Reconsideration and further examination are requested.

Documento 1
Respuesta de 12.462.795
Primera Declaración-Agosto 2013

REMARKS

A. Rejection of Claims 31 and 32 under 35 U.S.C. § 102(b)

Applicants acknowledge with appreciation the Examiner's withdrawal of the §102 rejection of Claims 31 and 32 over Backer *et al.* (U.S. 5,574,026).

B. Summary of Telephone Conference and Submission of Comparative Testing

The undersigned would like to thank Examiner Badio for the helpful discussion regarding further prosecution of the present application that occurred during a telephone conference between the undersigned and the Examiner on July 24, 2013. Following the teleconference, Applicants conducted head-to-head comparative testing of the sole compound recited in Claims 31and 32 of the present application (referred to herein as "TP-434"), and the compound of Example 5 of U.S. Patent No. 5,574,026 (the "Backer Compound"). The Examiner has relied on the Backer Compound in rejecting the pending claims and advised the undersigned during the referenced teleconference that she believes it is structurally the closest prior art compound. The results of the comparative testing are filed concurrently herewith in a Declaration of Joyce A. Sutcliffe, Ph.D. under 37 CFR §1.132 (the "Sutcliffe Declaration"), the Senior Vice President of Biology at Tetraphase Pharmaceuticals, Inc., the assignee of the present application (Sutcliffe Declaration, ¶1). As described in detail in the Sutcliffe Declaration, TP-434 has unexpectedly superior antibacterial activity in comparison to the Backer Compound.

C. Rejection of Claims 31 and 32 under 35 U.S.C. § 103(a)

The Examiner has maintained the rejection of Claims 31 and 32 under 35 U.S.C. § 103(a) over Backer *et al.* The Examiner's position is that the Backer Compound is both structurally the closest prior art compound and a proper lead compound. Based on this, the Examiner applied the two-part *Takeda* test of chemical obviousness[1] to arrive
at the conclusion that Claims 31 and 32 are unpatentable.

---

[1] According to the two-part test, the prior art must supply both (1) a reason or motivation to select *this* structurally close compound as a "lead compound" from among other prior art compounds; and (2) a reason or motivation to modify the *selected* lead compound to make the claimed compound with a reasonable expectation of success. *See Takeda Chem Ind. v. Alphapharm Pty., Ltd.*, 492 F.3d 1350, 1356 (Fed. Cir. 2007); *See Otsuka Pharm. Co. v. Sandoz, Inc.*, 678 F.3d 1280, 1291-1292 (Fed. Cir. 2012).

Applicants have set forth in detail in both their January 23, 2013 Pre-Appeal Brief Request for Review and their June 7, 2012 Reply the legal reasons why the Backer Compound is not a proper lead compound and why the *Takeda* two-part test for chemical obviousness was misapplied. Applicants continue to rely on those arguments to support their position that the Examiner has failed to demonstrate *prima facie* obviousness of the pending claims. However, it is not necessary to reiterate those legal arguments in this paper because the additional evidence presented herein demonstrates the non-obviousness of the claimed invention regardless of the correctness of the Examiner's obviousness rejection. This evidence unequivocally demonstrates that the antibacterial potency of the compound of Claims 31 and 32 is unexpectedly superior compared to the Backer Compound.

Under Dr. Sutcliffe's direction, comparative antibacterial testing of TP-434 and the Backer Compound was performed (Sutcliffe Declaration, ¶5). The *in vitro* testing reported in the Sutcliffe Declaration was conducted to determine the Minimum Inhibitory Concentration (MIC) values for TP-434 and the Backer Compound in 15 different bacterial strains (Sutcliffe Declaration, ¶5 and Appendix 5). Lower MIC values indicate more potent antibacterial activity (Sutcliffe Declaration, ¶6).

TP-434 and the Backer Compound were tested against the same diverse panel of bacteria used in Example 13 of the instant application (Sutcliffe Declaration, ¶8 and Appendix 5). The MIC values for each compound against each bacterial strain are shown in Appendix 5 of the Sutcliffe Declaration. The shaded areas of Appendix 5 highlight each instance where TP-434 was more potent than the Backer Compound, with the bottom row of the table in Appendix 5 providing the fold increase in potency of TP-434 over the Backer Compound (Sutcliffe Declaration, ¶8 and Appendix 5). In twelve of the fifteen bacterial strains tested, TP-434 was 2- to 8-fold more potent, and at least 4-fold more potent for the majority of those strains (Sutcliffe Declaration, ¶9 and Appendix 5). Based on these results, Dr. Sutcliffe opines that the antibacterial potency of TP-434 is unexpectedly superior when compared to the Backer Compound (Sutcliffe Declaration, ¶11). This data rebuts the obviousness rejection raised by the Examiner because even if the Backer Compound were a proper lead compound (which it is not), there was no reasonable expectation that changing its 7-position iodo substituent to a fluoro (as in TP-434) would result in a compound having more potent antibacterial activity (Sutcliffe

Declaration, ¶11). This unexpected increase in potency overcomes any *prima facie* obviousness of the claimed compound. *In re Chupp*, 816 F.2d 643, 646, 2 U.S.P.Q. 2d 1437, 1439 (Fed. Cir. 1987) and MPEP 716.02(a). Accordingly, withdrawal of the rejection is respectfully requested.

SUMMARY AND CONCLUSIONS

The subject matter of Claims 31 and 32 meets the requirements of 35 U.S.C. § 103(a) in view of Backer. The data presented in the Sutcliffe Declaration overcomes the Examiner's rejection regardless of the legal sufficiency of the *prima facie* obviousness rejection.

In view of the above, Applicants respectfully request reconsideration and withdrawal of this rejection. In view of the Request for Prioritized Examination being filed concurrently, Applicants respectfully request that the Examiner contact the undersigned before issuing the next Office Communication in an effort to expedite prosecution.

Respectfully submitted,
HAMILTON, BROOK, SMITH & REYNOLDS, P.C.

By Susan M. Abelleira

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Concord, MA 01742-9133
Date: August 23, 2013

1497774.1

SMA
August 23, 2013

PATENT APPLICATION
Attorney's Docket No.: 4142.1017-006

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Inventors:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	12/462,795	Group:	1628
Filed:	August 7, 2009	Examiner:	Badio, Barbara P.
Confirmation No:	5985		
For:	C7-Fluoro Substituted Tetracycline Compounds		

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DECLARATION OF JOYCE A. SUTCLIFFE, Ph.D. UNDER 37 CFR § 1.132

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Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

The undersigned, Joyce A. Sutcliffe, Ph.D., hereby declares that:

1. I am the Senior Vice President of Biology at Tetraphase Pharmaceuticals, Inc. (hereinafter "Tetraphase"), the assignee of the above-noted application. I have worked at Tetraphase since May 2009. I received my Ph.D. in microbiology from the University of Florida, Gainesville. After obtaining my degree, I held postdoctoral positions at the University of Massachusetts

Medical School and the National Institutes of Health. Prior to my employment by Tetraphase, I was Vice President, Research, at NanoBio Corporation, where I led antimicrobial, antiviral and antifungal discovery activities. In addition, I was Chief Research Scientist and Vice President, Biology, at Rib-X Pharmaceuticals from 2001 to 2007 and spent sixteen years at Pfizer conducting research in the area of antibiotic discovery and development.

2. My work at Tetraphase focuses on novel antibiotics effective against serious and life-threatening infections such as multidrug-resistant (MDR) infections. Developing such antibiotics is a pressing and unmet medical need, particularly for MDR Gram-negative bacteria (*see* Appendix 1). One such antibiotic I work on at Tetraphase is commonly referred to as TP-434 or eravacycline. The composition of matter for TP-434 and pharmaceutical compositions comprising TP-434 are the subject matter claimed in the above-noted application (the "Application").

3. TP-434 has broad spectrum antibacterial activity and has recently demonstrated a high clinical cure rate[1] against a wide variety of multidrug-resistant Gram-negative and Gram-positive aerobic and anaerobic bacteria in a Phase 2 human clinical trial (IV administration) for the treatment of patients with complicated intra-abdominal infections (cIAI) (*see* Appendix 2 for results). In addition, the FDA has designated TP-434 as a Qualified Infectious Disease Product (QIDP)[2] and the Infectious Diseases Society of America (IDSA) has identified TP-434 as one of only seven new drugs in development for the treatment of infections caused by Gram-negative bacteria (*see* Appendix 3).

4. As indicated in Paragraph 2 above, I am familiar with the Application and I am aware that in a Final Office Action issued from the U.S. Patent Office on July 24, 2012 (the "Final Office Action"), the Examiner rejected the claims as allegedly obvious under 35 U.S.C. §103(a) over Backer *et al.* (U.S. Patent No. 5,574,026). From my review of the Office Action, I understand the Examiner's rejection to be that the claimed compound (TP-434) and Example 5 of U.S. Patent No. 5,574,026 (the "Backer Compound") are structurally similar compounds and would

[1] Cure rate was based on the Microbiologically Evaluable (ME) population.

[2] QIDP designation was granted for treatment of complicated intra-abdominal infections (cIAI) and complicated urinary tract infections via IV administration.

be expected to have similar activity. Structural representations of TP-434 and the Backer Compound are presented in Appendix 4.

5. I make this declaration to present the results of *in vitro* testing performed at Tetraphase under my direction to determine the Minimum Inhibitory Concentration (MIC) values[3] for TP-434 and the Backer Compound against a variety of bacterial strains. I also make this declaration to show, based upon those results, that TP-434 has unexpectedly superior antibacterial activity in comparison to the Backer Compound.

6. The results presented in Appendix 5 were generated according to the Clinical and Laboratory Standards Institute (CLSI) performance standards using the MIC assay and bacterial strains set forth in Example 13 of the above-noted Application. The MIC values correlate with antibacterial activity, with a lower value being indicative of greater potency. For completeness, Appendix 6 describes the details of the strains used to generate the results reported herein, as well as an abbreviated description of the MIC assay used to compare the two compounds.

7. The Backer Compound was prepared following the procedures in U.S. Patent No. 5,574,026 to Backer *et al.* Details of the synthesis and characterization of the Backer Compound can be found in Appendix 7. TP-434 was prepared according to the procedure set forth in the specification of the above-noted application.

8. Appendix 5 compares the results of testing TP-434 and the Backer Compound against a diverse panel of bacteria that included both Gram-positive and Gram-negative bacteria. The panel of bacteria is the same as the panel used in Example 13 of the above-noted application. The shaded areas of Appendix 5 point to each instance where TP-434 is unexpectedly more potent than the Backer Compound. The bottom row of the table shown in Appendix 5 provides the fold increase in potency of TP-434 over the Backer Compound.

9. The results in Appendix 5 show that TP-434 demonstrates superior potency in comparison to the Backer compound in 12 of the 15 bacterial strains tested (see shaded areas). The fold

[3] Minimum Inhibitory Concentration (MIC) values are defined as the lowest concentration of an antimicrobial that will inhibit the visible growth of a microorganism after overnight incubation (18-24 hours at 35°C±2°C). MICs are used to determine the *in vitro* activity of antimicrobials with lower MICs values indicative of greater potency.

increase in potency corresponding to the 12 shaded areas in Appendix 5 range from a value of 2 to a value of 8, with the majority of the values indicating a 4 fold or more increase.[4] It is important to note that in all of the tetracycline resistant strains where a conclusion of relative potency can be made (strains indicated by an asterisk in Appendix 5), TP-434 is 4 to 8 fold more potent than the Backer Compound.

10. MIC values for both compounds against two of the remaining strains (SP106 and SP160) are at the lower numerical limit of the assay and no conclusion with respect to relative potency of TP-434 and the Backer Compound can be made. In the one remaining strain, TP-434 and the Backer Compound show equivalent potency.

11. Based on the results presented in Appendix 5 and discussed in Paragraph 9 above, it is my opinion that the antibacterial potency of TP-434 is unexpectedly superior when compared to the Backer Compound. This is because I would not have expected the replacement of the iodo substituent in the Backer Compound with the fluoro substituent of TP-434 to alter potency. The fact that TP-434 shows superior potency in 80% (12 of 15) of the bacterial strains tested and, to the extent discernible in the present assay, equivalence in the other three strains, is an unexpected result.

12. I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: 8/23/2013

Joyce A. Sutcliffe, Ph.D.

[4] In strains where the MIC of TP-434 was at or below the lowest numerical limit of the assay and the MIC of the Backer Compound was above this limit, the fold increase in indicated as "2 or more"; where the MIC of the Backer Compound was above the highest numerical limit of the assay and the MIC of TP-434 was below this limit, the fold increase is indicated as "more than 2."

APPENDIX 1: Hersh AL, Newland JG, Beekmann SE, Polgreen PM, Gilbert DN. 2012. Unmet medical need in infectious diseases. Clin Infect Dis 54:1677-1678.

APPENDIX 2: Poster LI-1647a, Presented at the 52nd Annual Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC), September 9-12, 2012, J. Solomkin, G. Cesnauskas, M. Ramesh, S. Walpole, J. Sutcliffe, P. Horn, "Efficacy and Safety of TP-434 (Eravacycline) versus Ertapenem in Complicated Intra-Abdominal Infection (cIAI)"

APPENDIX 3: Clinical Infectious Diseases (Advance Access, published April 17, 2013), Helen W. Boucher *et al.*, 10 x '20 Progress-Development of New Drugs Active Against Gram-Negative Bacilli: An Update From the Infectious Diseases Society of America"

APPENDIX 4: Structural Representations of TP-434 and Backer Compound

APPENDIX 5: Results of testing TP-434 and Backer Compound in MIC Assay.

APPENDIX 6: MIC Assay and Bacterial Strains

APPENDIX 7: Description of Synthesis used to Prepare Backer Compound

SMA/saj
November 25, 2013

PATENT APPLICATION
Attorney's Docket No.: 4142.1017-006
Expedited Procedure under 37 CFR § 1.116
Examining Group 1628

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	12/462,795	Group:	1628
Filed:	August 7, 2009	Examiner:	Badio, Barbara P.
Confirmation No:	5619		
For:	C7-Fluoro Substituted Tetracycline Compounds		

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REPLY AFTER FINAL OFFICE ACTION

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Commissioner:

This Reply is being filed in response to the Final Office Action mailed from the U.S. Patent and Trademark Office on September 23, 2013 in the above-identified application. Applicants are filing this Reply before the expiry of the 3-month statutory period (December 23, 2013) and within two months of the mailing date of the Final Office Action. No fees are believed due.

A *Second* Declaration under 35 U.S.C § 1.132 of Dr. Joyce Sutcliffe is being filed herewith. The Second Declaration expands on the scientific rationale supporting the unexpectedly superior activity of the claimed compound over the prior art compound. The Second Declaration could not have been presented earlier as it addresses statements by the Examiner presented in the record for the first time in the Final Office Action. Reconsideration and withdrawal of the pending rejection are requested.

Documento 2
4142.1017-006 AAF

REMARKS

I. Applicants' Claimed Compound Meets an Unmet Need

The U.S. Centers for Disease Control and Prevention ("CDC") recently estimated that more than two million people fall ill from antibiotic resistant bacteria every year in the United States, and that at least 23,000 die from these infections.[1] While resistance is escalating,[2] the development and approval of antibiotics has been steadily decreasing over the past three decades.[3] From 2010 through 2012, only *two* new antibiotics have been approved;[4] and only *a single* tetracycline antibiotic has been approved in the past 30 years. The Associate Director of the CDC recently declared:

> We're here. We're in the post-antibiotic era. There are patients for whom we have no therapy, and we are literally in a position of having a patient in a bed who has an infection, something that five years ago even we could have treated, but now we can't.[5]

Applicants' claimed compound is a broad spectrum antibiotic commonly referred as TP-434 or eravacycline. As stated by Dr. Sutcliffe in the Second Declaration of Joyce A. Sutcliffe, Ph.D., under 37 CFR §1.132 (the "Second Sutcliffe Declaration), submitted herewith, a first Phase 3 clinical trial of eravacycline is underway for the treatment of complicated intra-abdominal infection ("cIAI"), and a second Phase 3 trial for the treatment of complicated urinary tract infection ("cUTI") is planned to start before the end of 2013. Approval of eravacycline will provide a vitally important weapon in the war against the increasingly common and growing public health threat of antibiotic resistant infections. It is for this reason that the Food and Drug Administration ("FDA") designated the intravenous formulation of eravacycline as a "qualified infectious disease product" ("QIDP"), thereby making it eligible for, amongst other things, fast track designation and priority review by the FDA.

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[1] U.S. CENTERS FOR DISEASE CONTROL AND PREVENTION, U.S. DEPARTMENT OF HEALTH AND HUM AN SERVICES, ANTIBIOTIC RESISTANCE THREATS IN THE UNITED STATES, 2013, 11 *available* at http://www.cdc.gov/drugresistance/threat-report-2013/pdf/ar-threats-2013-508.pdf. (hereinafter "CDC REPORT")

[2] Adam L. Hersh et al., *Unmet Medical Need in Infectious Diseases*, 54 CLINICAL INFECTIOUS DISEASES 1677, 1677 (2012).

[3] CDC REPORT, *supra* note 1, at 44.

[4] *Id.*

[5] *Dr. Arjun Srinivasan: We've Reached "The End of Antibiotics, Period,"* *Frontline*, PUBLIC BROADCASTING SERVICE (October 22, 2013), http://www.pbs.org/wgbh/pages/frontline/health-science-technology/hunting-the-nightmare-bacteria/dr-arjun-srinivasan-weve-reached-the-end-of-antibiotics-period (edited transcript of interview with Dr. Arjun Srinivasan, Associate Director, U.S. Centers for Disease Control and Prevention (June 28, 2013)).

II. The First Sutcliffe Declaration

On August 23, 2013, Applicants filed the Declaration of Joyce A. Sutcliffe, Ph.D. Under 37 C.F.R. §1.132, hereinafter "the First Sutcliffe Declaration." In the First Sutcliffe Declaration, Applicants demonstrated that the claimed compound (TP-434) showed a 4-fold or more decrease in MIC values compared to the compound of Example 5 of U.S. Patent No. 5,574,026 to Backer *et al.* (the "Backer Compound") in a majority of the bacterial strains tested (First Sutcliffe Declaration, ¶9 and Appendix 5). Dr. Sutcliffe opined that such a decrease in MIC value was sufficiently substantial to translate into superiority in potency (First Sutcliffe Declaration, ¶9). Moreover, Dr. Sutcliffe found the increased potency to be unexpected given the 7-position iodo-to-fluoro change that differentiated the two compounds (First Sutcliffe Declaration, ¶11). Dr. Sutcliffe's opinion presented in the First Sutcliffe Declaration should have been sufficient to demonstrate non-obviousness under 35 U.S.C. § 103(a). As held by the court in *In re Soni* "when an applicant demonstrates substantially improved results, as Soni did here, and states that the results were unexpected, this should suffice to establish unexpected results in the absence of evidence to the contrary."[6]

Applicants respectfully submit that they have satisfied the test set forth in *In re Soni*. However, in contravention of the controlling law, the Examiner maintained the rejection of Claims 31 and 32 under 35 U.S.C. § 103(a) over U.S. Patent No. 5,574,026 ("Backer *et al.*"), on the grounds that unexpected results have not been, allegedly, demonstrated. In an effort to further explain why the MIC data presented in the First Sutcliffe Declaration demonstrates that the antibacterial activity of the claimed compound differs from the Backer Compound by such an appreciable degree as to be unexpected, Applicants submit herewith the Second Sutcliffe Declaration.

III. Rejection of Claims 31 and 32 Under 35 U.S.C. § 103(a)

The Examiner maintained the rejection of Claims 31 and 32 under 35 U.S.C. § 103(a) over U.S. Patent No. 5,574,026 ("Backer *et al.*"). The Examiner finds Applicants' showing of superior antibacterial potency of the presently-claimed compound relative to the Backer Compound (see the First Sutcliffe Declaration) insufficient to rebut the obviousness rejection of

[6] *In re Soni*, 54 F.3d 746, 751 (Fed. Cir. 1995).

A1

record. In particular, the Examiner stated that "[a]s shown by the Sutcliffe[ ] declaration both the exemplified prior art compound and the instantly claimed compound are broad spectrum antibacterial agents," "the showing of higher antibacterial activity in not unexpected since the skilled artisan in the art would expect differences in the degree of antibacterial property between the compounds," and "[d]etermining which compound is more potent would require only routine experimentation...."[7] The Examiner cites to MPEP § 716.02 and its summary of *In re Merck*[8]: "differences in sedative and anticholinergic effects between the prior art and claimed anti-depressants were not unexpected."

IV. Given a Presumption of Similar Properties for Similar Compositions, "Substantially Improved Properties" are *Ipso Facto* Unexpected

The Examiner's reliance on MPEP § 716.02 and its reference to *In re Merck* to support her assertion that differences in antibacterial property between the claimed invention and the prior art would be expected is misplaced.[9] The court in *In re Merck* did not hold that *all* differences in degree of a common property are expected. Specifically, the court in Merck found that there was "only a *slight* difference between the two compounds" [10] and that "*[i]n the absence of evidence* to show that the properties of the compounds differed in such an *appreciable degree* that the difference was really unexpected, we do not think that the Board erred in its determination that appellant's evidence was insufficient to rebut the prima facie case."[11] Therefore, the court in Merck found that differences in degree can be probative of non-obviousness when those differences are "appreciable."

Where evidence shows that the common properties of claimed and prior art compounds *do* differ in an appreciable degree, the question becomes whether that extent of the difference is unexpected. See also *In re Wagner*, 371 F.2d 877, 885 (C.C.P.A. 1967) (dismissing the BPAI's inference that a difference in degree cannot be used to demonstrate non-obviousness); and MPEP 716.02(a) ("Evidence of unobvious or unexpected advantageous properties, *such as superiority*

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[7] Final Office Action dated September 23, 2013, at page 4.
[8] *In re Merck & Co.*, 800 F.2d 1091, 1099 (Fed. Cir. 1986).
[9] Final Office Action at page 4 (citing *In re Merck & Co.*, 800 F.2d 1091 (Fed. Cir. 1986)).
[10] *In re Merck & Co.*, 800 F.2d 1091, 1099 (Fed. Cir. 1986) (emphasis added).
[11] *Id.*.

in a property the claimed compound shares with the prior art, can rebut prima facie obviousness.") (emphasis added).

In re Soni sets forth the legal basis for submission of comparative testing by applicants and the proper evaluation of such testing by the Office. As stated in *In re Soni*, "When a chemical composition is claimed, a *prima facie* case of obviousness under § 103 may be established by the PTO's citation of a reference to a similar composition, the presumption being that similar compositions have similar properties."[12] Therefore, the rejections of record must be based on this presumption.[13]

As the Federal Circuit held in *In re Soni*:

> One way for a patent applicant to rebut a prima facie case of obviousness is to make a showing of "unexpected results," *i.e.*, to show that the claimed invention exhibits some superior property or advantage that a person of ordinary skill in the relevant art would have found surprising or unexpected. The basic principle behind this rule is straightforward -- that which would have been surprising to a person of ordinary skill in a particular art would not have been obvious. The principle applies most often to the less predictable fields, such as chemistry, where minor changes in a product or process may yield substantially different results.[14]

Further, held the *In re Soni* Court,"[g]iven a presumption of similar properties for similar compositions, substantially improved properties are *ipso facto* unexpected."[15]

As explained in the Second Sutcliffe Declaration discussed in detail below, the evidence submitted by Applicants in the First Sutcliffe Declaration would lead one of ordinary skill in the art to conclude that TP-434 possesses substantially improved properties, compared to the Backer Compound. This is because the differences in antibacterial activity between TP-434 and the Backer Compound are sufficiently appreciable to consider further development of TP-434 as a broad spectrum antibacterial agent, while eliminating the Backer compound from consideration for further development as such an agent. This is both surprising and unexpected given the *expected* similarity in antibacterial activity. See the Second Sutcliffe Declaration, ¶ 7.

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[12] *In re Soni*, 54 F.3d 746, 750 (Fed. Cir. 1995).
[13] See, *e.g.*, Office Action mailed on January 24, 2012, at pp. 5-6 and the Final Office Action mailed on September 23, 2013, at p. 3.
[14] *In re Soni* at 750.
[15] *In re Soni* at 751.

V. The Second Sutcliffe Declaration

1. Differences in MIC Values of TP-434 and the Backer Compound Against E. coli Strain EC155 and K. pneumoniae Strain KP153 are Appreciable and Demonstrate a Substantially Improved Property, Unexpected in View of Structural Similarity

Referring to the Second Sutcliffe Declaration, ¶¶ 7-11, the demonstrated MIC values of the claimed compound (TP-434) against two clinically relevant, Gram-negative tetracycline resistant bacterial strains, *E. coli* strain EC155 and *K. pneumoniae* strain KP153, were sufficiently low to continue clinical development of the claimed compound as a broad spectrum antibiotic, while the Backer Compound MIC values against those same strains were too high to have considered that compound for further development as a broad spectrum antibiotic.

Specifically, as stated in the Second Sutcliffe Declaration, ¶10, the MIC values exhibited by the Backer Compound against Gram-negative bacterial strains EC155 and KP153 (8 μg/mL for each) are above the susceptibility (S) value for known tetracyclines (4 μg/mL) against this family of bacteria, while the MIC values for TP-434 against the same strains (1 μg/mL) are well below the susceptibility (S) value. As set forth in the Second Sutcliffe Declaration at ¶11, it is Dr. Sutcliffe's opinion that the failure of the Backer Compound to demonstrate a MIC value in the susceptibility range for known tetracyclines against EC155 and KP153 would disqualify it for further development as a broad spectrum antibiotic. In contrast, TP-434's MIC values against those strains are well within the susceptibility (S) range for known tetracyclines and would qualify it for continued development as a broad spectrum antibiotic, as now confirmed by the continued testing of TP-434 in late phase human clinical trials.

Thus, the data presented in the First Sutcliffe Declaration demonstrates such an appreciable difference in a common property of TP-434 and the Backer Compound, that it represents a substantially improved property of TP-434.

Dr. Sutcliffe further states in the Second Sutcliffe Declaration, ¶11, that prior to the head-to-head comparative testing of TP-434 and the Backer Compound, she could not have predicted that the MIC values against these strains would differentiate between TP-434 and the Backer Compound to such an extent that one would be selected for further development as a broad spectrum antibiotic and the other would not. Thus, the substantially improved property of TP-434 is unexpected given the structural similarity of the two compounds.

The difference in susceptibility in two important bacterial strains, by itself, is enough to demonstrate non-obviousness. "Evidence that a compound is unexpectedly superior in one of a spectrum of common properties ... can be enough to rebut a prima facie case of obviousness." *In re Chupp*, 816 F.2d 643, 646, 2 USPQ2d 1437, 1439 (Fed. Cir. 1987) (Evidence showing that the claimed herbicidal compound was more effective than the closest prior art compound in controlling quackgrass and yellow nutsedge weeds in corn and soybean crops was sufficient to overcome the rejection under 35 U.S.C. §103, even though the specification indicated the claimed compound was an average performer on crops other than corn and soybean). See also, *Ex parte A*, 17 USPQ2d 1716 (Bd. Pat. App. & Inter. 1990) (unexpected superior therapeutic activity of claimed compound against anaerobic bacteria was sufficient to rebut *prima facie* obviousness even though there was no evidence that the compound was effective against all bacteria); and MPEP 716.02(a) ("No set number of examples of superiority is required" to demonstrate non-obviousness).

In view of the above, the data presented in the First Sutcliffe Declaration and further explained in the Second Sutcliffe Declaration, ¶7-11, are sufficient to rebut the *prima facie* case of obviousness alleged by the Examiner over Backer *et al.*

2. *4- and 8-fold Improvements in the Potency of TP-434 Relative to the Backer Compound are Appreciable Differences and Represent Substantially Improved Properties, Unexpected in View of Structural Similarity*

Referring to the Second Sutcliffe Declaration, ¶¶12-15, the demonstrated and unexpected superiority of TP-434 over the Backer Compound against the majority of bacterial strains tested translates into therapeutic superiority in increased antibiotic resistance suppression and decreased risk of toxicity and other side effects.

Specifically, as stated in the Second Sutcliffe Declaration, ¶12, TP-434 showed at least a 4-fold decrease in MIC value in eight strains (six at 4-fold and two at 8-fold), as compared to the Backer Compound. In no strains tested was TP-434 less potent than the Backer Compound.

As explained by Dr. Sutcliffe in the Second Sutcliffe Declaration, ¶14, an important property of an antibiotic agent is its ability to suppress the emergence of resistant bacterial strains. Dr. Sutcliffe states that for two antibacterial agents similar in the limiting clinical factors such as toxicity and patient tolerance, the one exhibiting a lower MIC is likely to have an

advantageously higher potential for antibiotic resistance suppression. Dr. Sutcliffe further states that given the structural similarity of TP-434 and the Backer Compound, these compounds would be expected to display similarity in the limiting factors. Therefore, the lower MIC values exhibited by TP-434 are consistent with a higher potential for antibiotic resistance suppression. Thus, 4- and 8-fold improvements in the potency of TP-434 relative to the Backer Compound are appreciable differences and represent substantially improved properties.

Dr. Sutcliffe further states in the Second Sutcliffe Declaration, ¶15, that the results presented in the First Sutcliffe Declaration suggest that TP-434 could be administered at a lower dose than the Backer Compound to achieve the same therapeutic effect and thus demonstrate reduced side-effects and/or toxicity, yet another substantially improved property.

Referring to the Second Sutcliffe Declaration, ¶16, Dr. Sutcliffe concludes that the extent of the differences between TP-434 and the Backer Compound is surprising and unexpected.

VI. Summary and Conclusion

The legal presumption that similar compounds have similar properties can be rebutted by demonstrating that the claimed compound exhibits an appreciable difference in some property shared with the prior art, which results in a substantial improvement. Under the controlling law, substantially improved properties are *ipso facto* unexpected, and thus the compound possessing such substantially improved property is not obvious and meets the requirements of 35 U.S.C. § 103(a).

Based on the data presented and explained in the First and Second Sutcliffe Declarations, the antibacterial properties of TP-434 are substantially improved compared to those of the Backer Compound and unexpected, given the structural similarity. First, the differences between the MIC values of TP-434 and the Backer Compound for certain clinically important strains are so appreciable that a person of ordinary skill in the art would select TP-434, but not the Backer Compound, for further development as a broad spectrum antibiotic. Second, the 4- and 8-fold improvements in MIC values demonstrated by TP-434 translate into superior resistance suppression and decreased risk of side effects and/or toxicity.

In view of the above, Applicants respectfully request reconsideration and withdrawal of the rejection. It is believed that all claims are in condition for allowance and it is respectfully requested that the application be passed to issue. If the Examiner feels that a telephone conference would expedite prosecution of this case, the Examiner is invited to call the undersigned.

Respectfully submitted,

HAMILTON, BROOK, SMITH & REYNOLDS, P.C.

By Susan M. Abelleira

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Concord, MA 01742-9133

Date: November 25, 2013

SMA/AA
November 24, 2013

PATENT APPLICATION
Attorney's Docket No.: 4142.1017-006

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Inventors:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	12/462,795	Group:	1628
Filed:	August 7, 2009	Examiner:	Badio, Barbara P.
Confirmation No:	5985		
For:	C7-Fluoro Substituted Tetracycline Compounds		

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SECOND DECLARATION OF JOYCE A. SUTCLIFFE, Ph.D., UNDER 37 CFR § 1.132

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Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

The undersigned, Joyce A. Sutcliffe, Ph.D., hereby declares that:

1. My background and present position are set forth in my previous declaration filed on August 23, 2013 in this application ("the First Sutcliffe Declaration").

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2. The background and demonstrated efficacy of TP-434, the subject compound of this application, is also set forth in the First Sutcliffe Declaration. Since that submission, TP-434 has entered a Phase 3 clinical trial for the treatment of complicated intra-abdominal infection ("cIAI"). Also, a second Phase 3 trial for the treatment of complicated urinary tract infection ("cUTI") is now planned to begin before the end of 2013.

3. I am familiar with the above Application and I am aware that in a Final Office Action dated September 23, 2013 (the "Office Action"), the Examiner continued to reject the claims as obvious under 35 U.S.C. §103(a) over Backer *et al.* (U.S. Patent No. 5,574,026). I understand the Examiner is not convinced that the higher (i.e., more potent) antibacterial activity of the claimed compound ("TP-434") relative to the Example 5 compound of Backer (the "Backer Compound") demonstrated in the First Sutcliffe Declaration is unexpected.

4. I make this declaration to expand on the basis for my opinion, presented in the First Sutcliffe Declaration, of unexpectedly superior antibacterial activity of TP-434 in comparison to the Backer Compound. Specifically, I make this declaration to present additional scientific reasoning to support my opinion that the differences between the two compounds are appreciable and result in substantially improved properties.

5. Structural representations of TP-434 and the Backer Compound are presented in Appendix 4 of the First Sutcliffe Declaration. Reference to the comparative minimal inhibitory concentration (MIC)[1] data of Appendix 5 is to the data found in Appendix 5 of the First Sutcliffe Declaration listing MIC values of TP-434 and the Backer Compound against 15 bacterial strains.

6. I am informed and believe that the legal standard for establishing chemical obviousness based on structural similarity relies on the presumption that similar compounds have similar activities. Prior to obtaining the data presented in the First Sutcliffe Declaration, I was not aware of any information that suggested that this presumption should not be true for TP-434

[1] Minimum Inhibitory Concentration (MIC) values are defined as the lowest concentration of an antimicrobial that will inhibit the visible growth of a microorganism after overnight incubation (18-24 hours at 35°C±2°C). MICs are used to determine the *in vitro* activity of antimicrobials with lower MICs values indicative of greater potency.

and the Backer Compound. Accordingly, I have adopted this legal standard throughout this and my previous declarations.

7. In my opinion, the differences in antibacterial activity between TP-434 and the Backer Compound demonstrated in Appendix 5 are sufficiently appreciable to continue further development of TP-434 as a broad spectrum antibacterial agent, while eliminating the Backer compound from consideration for further development. This is both surprising and unexpected given the expected similarity in antibacterial activity. The reason for my opinion is set forth below.

8. MIC values are used by clinicians treating patients with bacterial infections to determine the proper choice of drug to be administered. The MIC value of a candidate antibiotic for treatment is determined against isolates of a patient's bacterium and then compared to standardized MIC values for that antibiotic as published by the Clinical and Laboratory Standards Institute ("CLSI"). The MIC values in the CLSI guidance[2] help guide the physician as to expected therapeutic outcome. Comparison to the standardized values (commonly referred to as breakpoints) permits evaluation of whether a patient's bacterium would be "Susceptible" (S), "Resistant" (R) to the tested antibiotic, or somewhere in between ("Intermediate" (I)). MIC values at or below the *Susceptible* value mean the patient's isolates are inhibited by a standard dose of that antibiotic, implying that it can be used with confidence to treat the patient's infection. MIC values at or above the *Resistant* value mean that the bacteria are not inhibited by the standard dose of the antibiotic, thus eliminating it as a choice for treatment. MIC values in the *Intermediate* range mean that the efficacy of the antibiotic against the patient's infection is suboptimal. An *Intermediate* value suggests use only where a dose higher than the standard can be administered or where the infection is located in body sites where the antibiotic will be physiologically concentrated.[3] To me an *Intermediate* value suggests seeking a more potent alternative antibiotic for treatment if one is available.

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[2] *See* CLINICAL AND LABORATORY STANDARDS INSTITUTE, PERFORMANCE STANDARDS FOR ANTIMICROBIAL SUSCEPTIBILITY TESTING; TWENTY-SECOND INFORMATION SUPPLEMENT, M100-S22, Vol. 32, No. 3 (2012).
[3] PERFORMANCE STANDARDS FOR ANTIMICROBIAL SUSCEPTIBILITY TESTING at 26.

9. Both TP-434 and the Backer Compound are tetracycline-type compounds. Therefore, the MIC values of Appendix 5 can be compared to standard MIC values of known tetracyclines to interpret the results. Exhibit 2-1 is an excerpt from "PERFORMANCE STANDARDS FOR ANTIMICROBIAL SUSCEPTIBILITY TESTING," published by the CLSI.[4] At page 47 of Exhibit 2-1, Interpretive Criteria for three approved tetracycline antibiotics (i.e, Tetracycline, Doxycycline and Minocycline) against bacteria of the *Enterobacteriaceae* family are reported. *Enterobacteriaceae* include the species *E. coli* and *K. pneumoniae*. For each of the three tetracyclines, susceptibility (S) is indicated by a MIC value of 4 μg/mL or less.

10. The shaded areas in Appendix 5 represent the 12 strains in which TP-434 demonstrated lower MIC values (increased antibacterial potency) in comparison to the Backer Compound. Of particular clinical significance are the results for the Gram-negative tetracycline-resistant strains, *E. coli* strain EC155 and *K. pneumoniae* strain KP153.[5] These two species are frequently isolated pathogens from patients with serious infections and are often multidrug-resistant and hard to eradicate. The resistance of these strains to most treatments supports the medical community's view that effective treatment of Gram-negative resistant bacterial infections is a pressing and unmet medical need.[6] The MIC values exhibited by the Backer Compound against representative *E. coli* strain EC155 and *K. pneumoniae* KP153 (8 μg/mL for each) are above the susceptibility (S) value for known tetracyclines (4 μg/mL) against this family of bacteria, while the MIC values for TP-434 against the same strains (1 μg/mL) are well below the susceptibility (S) value. These differences are both appreciable and unexpected.

11. In the clinical development of a new tetracycline, it is imperative that such compound is potent against strains expressing tetracycline-resistant mechanisms since these mechanisms are widespread and would render the compound unusable in the clinic for empiric therapy. MIC assays like the one used to generate the data in Appendix 5 are typically used as early

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[4] PERFORMANCE STANDARDS FOR ANTIMICROBIAL SUSCEPTIBILITY TESTING pp. 1-50.
[5] The designation *tet(A)* refers to the mechanism of tetracycline resistance and is further explained in Appendix 6 of the First Sutcliffe Declaration.
[6] Hersh, AL Newland JG, Beekmann SE, Polgreen PM, Gilbert DN, 2012. Unmet medical need in infectious diseases. Clin Infect Dis 54:1677-1678.

screening tools in the development of new antibiotics. In my view, the failure of the Backer Compound to demonstrate a MIC value in the susceptible (S) range for known tetracyclines against strains EC155 and KP153 would disqualify it for further development as an antibiotic. In contrast, TP-434's MIC values against those strains are well within the susceptible range for known tetracyclines and would qualify it for continued development, as now confirmed by the continued testing of TP-434 in late phase human clinical trials. Prior to testing TP-434 and the Backer Compound head-to-head, I could not have predicted that the MIC values against these strains would differentiate between TP-434 and the Backer Compound to such an extent that one would be selected for further development and the other would not. This substantially improved property of TP-434 is unexpected given the structural similarity of the two compounds.

12. The data in Appendix 5 also show that for the ten bacterial strains for which a definitive difference in MIC value could be determined[7], TP-434 showed at least a 4-fold decrease in MIC value in eight strains (six at 4-fold and two at 8-fold), as compared to the Backer Compound. Further, in no strain tested was TP-434 less potent than the Backer Compound.

13. The 4- and 8-fold improvements in the potency of TP-434 relative to the Backer Compound are appreciable differences. This is because of at least two important advantages associated with a lower MIC value -- increased antibiotic resistance suppression and decreased risk of side effects and/or toxicity.

14. Resistance suppression is accomplished by administering to a patient a concentration of antibiotic sufficient to eliminate, within certain statistical confidence, all infectious agents from the site of infection during the course of treatment (the "mutant prevention concentration"). Such elimination ensures that any strain with a mutation that confers antibiotic resistance does not survive and propagate, leading to an infectious population that is no longer susceptible to the antibiotic. The mutant prevention concentration is measured in multiples of the MIC, and the higher the multiple that can be achieved, the more statistically confident a practitioner can be of eliminating all infectious agents. However, the mutant

[7] Fifteen bacterial strains were tested. In five of the strains the exact fold difference could not be determined because the MIC values were either above or below the experimental limits.

prevention concentration is limited by clinical factors such as the toxicity and patient tolerance. Thus, for two antibacterial agents similar in these limiting clinical factors, the one exhibiting a lower MIC is likely to have an advantageously higher potential for antibiotic resistance suppression. Given the structural similarity of TP-434 and the Backer Compound, these compounds would be expected to display similarity in the limiting factors. Therefore, the lower MIC values exhibited by TP-434 are consistent with it having a higher potential for antibiotic resistance suppression. This is another substantially improved property of TP-434 over the Backer Compound.

15. Toxicity and other undesired side effects of an antibiotic agent are directly related to dose. An antibacterial agent exhibiting a lower MIC value has the potential to be administered at a lower dose to achieve a desired therapeutic effect, thus decreasing toxicity and other side effects. The results in Appendix 5 suggest that TP-434 could be administered in a lower dose than the Backer Compound to achieve the same therapeutic effect and thus demonstrate reduced side-effects and/or toxicity. Again, such difference is a substantially improved property of TP-434 over the Backer Compound.

16. Based on the above, it is my opinion that the antibacterial properties of TP-434 are substantially improved compared to those of the Backer Compound. This is because the differences between the MIC values of TP-434 and the Backer Compound for certain clinically important strains are so appreciable that I would select TP-434, but not the Backer Compound, for further development as an antibacterial agent. It is also because the 4- and 8-fold improvements in MIC demonstrated by TP-434 translate to superior resistance suppression and decreased risk of side effects and/or toxicity. Furthermore, given the legal presumption that structurally similar compounds have similar biological properties, I find the extent of the differences surprising and unexpected.

17. I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United

States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: 11/25/2013

Joyce A. Sutcliffe

Joyce A. Sutcliffe, Ph.D.

EXHIBIT 2-1: CLINICAL AND LABORATORY STANDARDS INSTITUTE, PERFORMANCE STANDARDS FOR ANTIMICROBIAL SUSCEPTIBILITY TESTING; TWENTY-SECOND INFORMATION SUPPLEMENT, M100-S22, Vol. 32, No. 3 (2012), p. 1-50.

PATENT APPLICATION
Attorney's Docket No.: 4142.1017-024

SMA
March 21, 2014

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Inventors:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	13/718,909	Group:	1628
Filed:	December 18, 2012	Examiner:	Badio, Barbara P.
Confirmation No:	5985		
For:	C7-Fluoro Substituted Tetracycline Compounds		

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REPLY

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Commissioner:

This Reply and Request for Continued Examination-Prioritized Examination under 37 C.F.R. §1.102(e)(2) are being filed in lieu of filing a Brief on Appeal, pursuant to the Notice of Appeal received in the U.S. Patent and Trademark Office on January 8, 2014 in the above-identified application. An extension of time to file the Brief on Appeal is respectfully requested. A Petition for Extension of Time and the appropriate fee are being filed concurrently.

Reconsideration and further examination are requested.

1601656.v1

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Documento 3
Declaración Combinada
1 y 2 en el caso de método.

REMARKS

Personal Interview-March 6, 2014

Applicants' Attorneys, Susan M. Abelleira and N. Scott Pierce, would like to thank Examiner Badio and Supervisory Examiners Shen and Celsa for granting the personal interview conducted on March 6, 2014 at the United States Patent & Trademark Office to discuss the instant application. Dr. Joyce Sutcliffe, Senior Vice President of Biology at Tetraphase Pharmaceuticals, Inc. (Assignee of the instant application), also attended and participated in the personal interview.

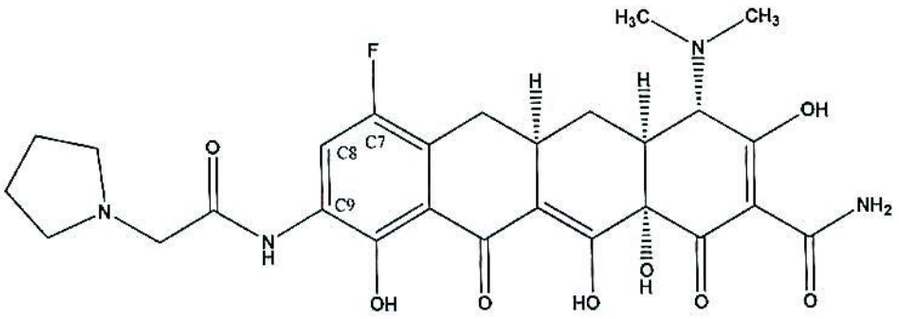
During the interview, it was agreed that submission of the declaratory evidence of record in related U.S. Application No. 12/462,795[1] would serve to overcome the obviousness rejections currently of record in the instant application. Applicants are submitting herewith copies of the First and Second Sutcliffe Declarations. In addition, to ensure completeness of the record Applicants are also presenting arguments addressing the outstanding rejections of record (See Final Office Action dated July 9, 2013).

The remainder of this Reply is set forth under the appropriate subheadings for the convenience of the Examiner.

Claim Status

Claims 1-16 are pending. Claim 1 is the sole independent claim and is directed to a method for reducing infections caused by bacteria in a subject in need thereof, comprising administering to said subject an effective amount of a compound represented by the following structural formula (or a pharmaceutically acceptable salt thereof):

[1] Declaration of Joyce A. Sutcliffe, Ph.D. Under 37 C.F.R. §1.132, filed at the USPTO on August 23, 2013 ("The First Sutcliffe Declaration"); and Second Declaration of Joyce A. Sutcliffe, Ph.D. Under 37 C.F.R. §1.132, filed at the USPTO on November 25, 2013 ("The Second Sutcliffe Declaration").



Compound 34 (a.k.a., TP-434 and Eravacycline).

Claim rejections under 35 U.S.C. §112 have been withdrawn. Claim rejections under 35 U.S.C. §103(a) are maintained.

Summary of Claim Rejections Under 35 U.S.C. §103

The Examiner maintained three separate grounds for rejection.

I. Rejection Over Backer

The Examiner maintained the previous rejection of the pending claims as being unpatentable over U.S. 5,574,026 to Backer *et al.* ("Backer") and, specifically, over the compound of Example 5 of Backer. (See, in particular, page 6 of the Office Action mailed February 5, 2013). In the most recent Office Action, the Examiner stated that she agrees that Backer does not teach the antibacterial properties of the compounds. The Examiner further stated, however, that tetracyclines, as a class, are widely used as antibiotics. As such, according to the Examiner, "even with the absence of antibacterial property of the compounds of Backer, based on the fact that compounds of Backer are tetracyclines, the skilled artisan in the art would have the reasonable expectation that they would have antibacterial property." The Examiner also stated that, "tetracyclines are well known antibacterial agent and, thus, the skilled artisan would have the reasonable [expectation] that selection of any of the prior art tetracycline compounds, including the modification of the exemplified compound by replacing a 7-iodo group with a 7-fluoro group would result in a tetracycline compound with antibacterial property."(Office Action, pp. 3-4.)

II. Rejection Over Sum in View of Bandarage

The Examiner maintained the rejection of pending claims as being obvious over EP 0 582 789 ("Sum") in view of U.S. Pat. No. 7,825,105 ("Bandarage"). In particular, the Examiner reasserted that Sum teaches an "8-halo" compound and that although Sum does not exemplify an "8-fluoro" compound, such a compound is encompassed by the genus taught by Sum, and is a "positional isomer" of Applicants' claimed compound.

III. Rejection over Backer, Sum and Bandarage in Combination

The Examiner rejected Claims 1-16 under 35 U.S.C. §103(a) as being unpatentable over Backer, Sum, and Bandarage, in combination. The Examiner stated that Backer teaches "7-halo compound [sic] inclusive of the claimed compounds," i.e. a chemical genus that includes the claimed compound, that Sum teaches a genus of 8-substituted tetracyclines, and that Bandarage teaches that both 7-halo and 8-halo-substituted tetracyclines have antibacterial properties. (Office Action, p. 9). The Examiner concludes that in combination, the cited prior art makes obvious the antibacterial property of 7-halo tetracycline derivatives, including the 7-fluoro compound.

Applicant's Response to Maintained Obviousness Rejections

I. Results of Applicants' Comparative Data Demonstrates Unexpected Results for TP-434

Applicants have set forth in detail below and in their May 5, 2013 Amendment the reasons why the Examiner has not made out a legally sufficient *prima facie* case of obviousness under 35 U.S.C. §103. Applicants do not concede that a *prima facie* case of obviousness has been established. As agreed during the personal interview conducted at the U.S. PTO on March 6, 2014, however, Applicants are submitting experimental data presented in the First Sutcliffe Declaration and further explained in the Second Sutcliffe Declaration submitted in the companion case, U.S. Application No. 12/462,795. It is Applicants' understanding that submission of this declaratory evidence will overcome the obviousness rejections currently of record in the instant application. Submitted herewith as **Exhibit A** and **Exhibit B** are copies of

the First and Second Sutcliffe Declarations, respectively. A discussion of the First and Second Sutcliffe Declarations follows.

In re Soni sets forth the legal basis for submission of comparative testing by Applicants and the proper evaluation of such testing by the Office. As the Federal Circuit held in *In re Soni*:

> One way for a patent applicant to rebut a prima facie case of obviousness is to make a showing of "unexpected results," *i.e.*, to show that the claimed invention exhibits some superior property or advantage that a person of ordinary skill in the relevant art would have found surprising or unexpected. The basic principle behind this rule is straightforward -- that which would have been surprising to a person of ordinary skill in a particular art would not have been obvious. The principle applies most often to the less predictable fields, such as chemistry, where minor changes in a product or process may yield substantially different results.

In re Soni, 54 F.3d 746, 750 (Fed. Cir. 1995).

The court in *Soni* further stated that,"[g]iven a presumption of similar properties for similar compositions, substantially improved properties are *ipso facto* unexpected." *Id.* at 751.

The evidence submitted by Applicants in the First Sutcliffe Declaration and further explained in the Second Sutcliffe Declaration would lead one of ordinary skill in the art to conclude that TP-434 possesses substantially improved properties compared to the compound of Example 5 of Backer. This is because the differences in antibacterial activity between TP-434 and the compound of Example 5 of Backer are sufficiently appreciable to consider TP-434 as a candidate for further development as a broad spectrum antibacterial agent, while eliminating the compound of Example 5 of Backer from such consideration. According to Dr. Sutcliffe, this is both surprising and unexpected given the general presumption of similarity in antibacterial activity based on structural similarity of the two compounds. Thus, the data presented in the First Sutcliffe Declaration and further explained in the Second Sutcliffe Declaration demonstrates *a substantially improved property* of TP-434 relative to the compound of Example 5 of Backer.

Referring to the Second Sutcliffe Declaration, ¶¶ 7-11, the demonstrated MIC values of the claimed compound (TP-434) against two clinically relevant, Gram-negative tetracycline

resistant bacterial strains, *E. coli* strain EC155 and *K. pneumonia* strain KP153, were sufficiently low to continue clinical development of the claimed compound as a broad spectrum antibiotic. In contrast, the values obtained for Compound 5 of Backer against those same strains were too high to have considered that compound for further development as a broad spectrum antibiotic. Specifically, as stated in the Second Sutcliffe Declaration, ¶10, the MIC values exhibited by the Compound 5 of Backer against Gram-negative bacterial strains EC155 and KP153 (8 μg/mL for each) are above the susceptibility (S) value for known tetracyclines (4 μg/mL) against this family of bacteria, while the MIC values for TP-434 against the same strains (1 μg/mL) are well below that susceptibility (S) value. As set forth in the Second Sutcliffe Declaration at ¶11, it is Dr. Sutcliffe's opinion that the failure of Compound 5 of Backer to demonstrate a MIC value in the susceptibility range for known tetracyclines against EC155 and KP153 would disqualify it for further development as a broad spectrum antibiotic. In contrast, TP-434's MIC values against those strains are well within the susceptibility (S) range for known tetracyclines and would qualify it for continued development as a broad spectrum antibiotic, as now confirmed by the continued testing of TP-434 in late phase human clinical trials.

Dr. Sutcliffe further states in the Second Sutcliffe Declaration, ¶11, that prior to the head-to-head comparative testing of TP-434 and the Backer Compound, she could not have predicted that the MIC values against these strains would differentiate between TP-434 and Compound 5 of Backer to such an extent that one would be selected for further development as a broad spectrum antibiotic and the other would not. Thus, the substantially improved property of TP-434 is unexpected given the structural similarity of the two compounds.

The difference in susceptibility in two important bacterial strains, by itself, is enough to demonstrate non-obviousness. "Evidence that a compound is unexpectedly superior in one of a spectrum of common properties ... can be enough to rebut a prima facie case of obviousness." *In re Chupp*, 816 F.2d 643, 646, 2 USPQ2d 1437, 1439 (Fed. Cir. 1987) (Evidence showing that the claimed herbicidal compound was more effective than the closest prior art compound in controlling quackgrass and yellow nutsedge weeds in corn and soybean crops was sufficient to overcome the rejection under 35 U.S.C. §103, even though the specification indicated the claimed compound was an average performer on crops other than corn and soybean). See also, *Ex parte A*, 17 USPQ2d 1716 (Bd. Pat. App. & Inter. 1990) (unexpected superior therapeutic

activity of claimed compound against anaerobic bacteria was sufficient to rebut *prima facie* obviousness even though there was no evidence that the compound was effective against all bacteria); and MPEP 716.02(a) ("No set number of examples of superiority is required" to demonstrate non-obviousness).

Again referring to the Second Sutcliffe Declaration, ¶¶12-15, the demonstrated and unexpected superiority of TP-434 over Compound 5 of Backer against the majority of bacterial strains tested translates into therapeutic superiority in increased antibiotic resistance suppression and decreased risk of toxicity and other side effects. Specifically, as stated in the Second Sutcliffe Declaration, ¶12, TP-434 showed at least a 4-fold decrease in MIC value in eight strains (six at 4-fold and two at 8-fold), as compared to the compound of Example 5 of Backer. In no strains tested was TP-434 less potent than the compound of Example 5 of Backer. As explained by Dr. Sutcliffe in the Second Sutcliffe Declaration, ¶14, an important property of an antibiotic agent is its ability to suppress the emergence of resistant bacterial strains. Dr. Sutcliffe states that, for two antibacterial agents similar in limiting clinical factors, such as toxicity and patient tolerance, the one exhibiting a lower MIC is likely to have an advantageously higher potential for antibiotic resistance suppression. Dr. Sutcliffe further states that, given the structural similarity of TP-434 and the compound of Example 5 of Backer, these compounds would be expected to display similarity in those limiting factors. Therefore, the lower MIC values exhibited by TP-434 are consistent with a higher potential for antibiotic resistance suppression. Thus, 4- and 8-fold improvements in the potency of TP-434 relative to the compound of Example 5 of Backer are appreciable differences and represent substantially improved properties.

Dr. Sutcliffe further states in the Second Sutcliffe Declaration, ¶15, that the results presented in the First Sutcliffe Declaration suggest that TP-434 could be administered at a lower dose than the compound of Example 5 of Backer to achieve the same therapeutic effect and thus demonstrate reduced side-effects and/or toxicity, yet another substantially improved property. Referring to the Second Sutcliffe Declaration, ¶16, Dr. Sutcliffe concludes that the extent of the differences between TP-434 and the compound of Example 5 of Backer is surprising and unexpected.

In view of the declaratory evidence discussed above, Applicants have rebutted any *prima facie* case of obviousness by presentation of evidence demonstrating an appreciable difference

and substantially improved properties of TP-434 over the compound of Example 5 of Backer. Absent evidence to the contrary, the demonstrated substantially improved properties of TP-434 are *ipso facto* unexpected, thereby rebutting any *prima facie* case of obviousness as a matter of law. Therefore, Applicants' Claim 1 and claims dependent thereon meet the requirement of 35 U.S.C. §103(a), and Applicants respectfully request reconsideration and withdrawal of all outstanding rejections of these claims.

II. The Law of Chemical Obviousness

Establishing a *prima facie* case of chemical obviousness must follow the well-established legal precedents that emphasize the need to avoid impermissible hindsight. Such hindsight is avoided by requiring that the Examiner, without the benefit of knowing the claimed structure, find guidance in the prior art to select an appropriate lead chemical compound from the prior art and to modify its chemical structure in a manner necessary to arrive at the claimed invention.

The issue of chemical obviousness was most recently revisited by the Federal Circuit in *Pfizer, Inc. v., TEVA Pharm. USA, Inc.*, 2012-1576, -1601, -1602, -1603, -1604, -1605, -1607 (February 6, 2014) (non-precedential). The court summarized that under its precedent, a *prima facie* case of obviousness "under our precedent" is a two-part analysis of: 1) lead compound selection that extends beyond "mere structural similarity"; and 2) whether the prior art would have taught "specific molecular modifications" to the selected lead compound:

> Whether a new chemical compound would have been prima facie obvious over particular prior art compounds follows a two-part inquiry under our precedent. First, the court determines whether a chemist of ordinary skill in the art would have selected the asserted prior art compound as a lead compound, or starting point, for further development. *Eisai Co. v. Dr. Reddy's Labs., Ltd.*, 533 F.3d 1353, 1359 (Fed. Cir. 2008). A lead compound is a compound in the prior art that would be "most promising to modify in order to improve upon its activity and obtain a compound with better activity." *Takeda Chem, Indus., Ltd. v. Alphpharm Pty., Ltd.*, 492 F.3d 1350, 1357 (Fed. Cir. 2007). The selection analysis may be guided by evidence of the compound's pertinent properties, such as chemical activity or potency. *See Eli Lilly & Co. v. Zenith Goldline Pharm., Inc.*, 471 F.3d 1369, 1378 (Fed. Cir. 2006). Mere structural similarity between a prior art compound and the claimed compound does not form the lead compound selection. *Otsuka Pharm. Co. v. Sandoz Inc.*, 678 F.3d

> 1280, 1292 (Fed. Cir. 2012); *see Daichii Sankyo Co. v. Matrix Labs., Ltd.*, 619 F.3d 1346, 1354 (Fed. Cir. 2010).
>
> ***
>
> A patent challenger, however, must demonstrate the selection of a lead compound based on its "promising useful properties," not a hindsight-driven search for structurally similar compounds. *Daichii,* 619 F.3d at 1354.
>
> ***
>
> The second step of the obviousness analysis requires a showing that the prior art would have taught a skilled artisan to make "specific molecular modifications" to a lead compound so that the claimed compound may be made with a reasonable expectation of success. *Id.* at 1356-57.
>
> ***
>
> Indeed, a vague suggestion in the prior art pointing to a broad class of compounds, is not a teaching of "specific molecular modification" required by our precedent. *Takeda,* 492 F.3d at 1356.

Once a structurally close compound is found, it must then pass a two-part test. Finding the structurally similar compound is a prerequisite to, but not a part of, the two-part test. According to the two-part test, the prior art must supply both (1) a reason or motivation to select *this* structurally close compound as a "lead compound" from among other prior art compounds; and (2) a reason or motivation to modify the *selected* lead compound to make the claimed compound with a reasonable expectation of success. *See Takeda Chem Ind. v. Alphapharm Pty., Ltd.*, 492 F.3d 1350, 1356 (Fed. Cir. 2007); *See Otsuka Pharm. Co. v. Sandoz, Inc.*, 678 F.3d 1280, 1291-1292 (Fed. Cir. 2012), *cert. denied*, 2013 U.S. Lexis 891 (U.S. Jan. 14, 2013). These two inquiries of the test articulated in *Takeda* are independent of each other; both must be satisfied in order to build a *prima facie* case of chemical obviousness.

As stated in *Eisai Co. Ltd. v. Dr. Reddy's Laboratories Ltd.*, 533 F.3d 1353 (Fed. Cir. 2008), "[o]bviousness based on structural similarity thus can be proved by identification of some motivation that would have led one of ordinary skill in the art to select and then modify a known

compound (i.e. a lead compound) in a particular way to achieve the claimed compound." *Eisai* at 1357. As can be seen, motivation to select "a known compound" is required. In *Otsuka Pharm. Co. v. Sandoz, Inc.*, 678 F.3d 1280, 1291-1292 (Fed. Cir. 2012), the Federal Circuit has reiterated that "[a] lead compound, as we have explained, is 'a compound in the prior art that would be most promising to modify in order to improve upon its ... activity and obtain a compound with better activity.'" *Otsuka* at 1291 citing *Takeda Chem Ind. v. Alphapharm Pty., Ltd.*, 492 F.3d 1350, 1357 (Fed. Cir. 2007). This explanation by the Federal Circuit makes it clear that the first part of the two-part test of *Takeda* -- motivation to select a prior art compound -- turns on the properties of the proposed lead compound; structural similarity is not enough.

A Case Law chart is provided in Exhibit C, that summarizes certain of the leading Federal Circuit cases that address the selection of a lead compound. Supporting citations are provided in Exhibit D. The chart includes three columns. For each case, the shorthand case name is in the left column, the center column reports whether there was sufficient legal basis to select the "proposed" lead compound for further development (i.e., whether the proposed lead passed the first part of the two part *Takeda* test) and the right column summarizes the rationale offered by the Federal Circuit.

The Case Law chart clearly shows that structural similarity alone is not sufficient to complete a reasoned identification of a "lead compound." If structural similarity alone were enough, then the middle column of the chart would be **YES** in each instance, because all of the proposed lead compounds were selected based on structural similarity with the claimed compound. Also, if structural similarity alone were sufficient, then the pertinent properties of the proposed lead prior art compounds would be irrelevant to identification of a lead compound determination, rather than informing that identification, as noted in the chart. Applicants point out that the chart includes many of the cases referred to in the *2010 KSR Guidelines Update* from the *Federal Register* (*Federal Register*, Vol 75, No 169, pp 53651-652).

Federal Circuit case law requires that evidence of all of a prior art compound's pertinent properties should guide its selection as a lead compound. Such properties may include positive properties, such as activity and potency; adverse effects, such as toxicity; and other relevant characteristics in evidence. *See Otsuka* at 1292. "Absent a reason or motivation based on such prior art evidence, mere structural similarity between a prior art compound and the claimed

compound does not inform the lead compound selection." *Id.* at 1292. The table of Exhibit C shows that in each case, the properties of a structurally similar proposed lead compound relative to other prior art compounds (e.g., activity, toxicity and side effects, etc.) was a key factor in whether it met the legal standard as a lead.

For example, in *Takeda*, the prior art "compound b" was not considered a lead compound because it was known to have serious side effects that would be deleterious for its intended purpose. "[C]ompound b was not identified as one of the three most favorable compounds" in terms of toxicity and activity identified in one of the prior art documents (Sodha II). *Takeda* at 1358.

In *Daiichi Sankyo Co., Ltd. v. Matrix Labs., Ltd.*, 619 F.3d 1346 (Fed. Cir. 2010) six compounds disclosed as angiotensin receptor blockers (ARBs) in the '902 prior art patent ("the '902 ARBs") were the closest structurally to the active ingredient of the patented prodrug compound olmesartan medoxamil. One of those six compounds differed from the active ingredient by just one atom (*i.e.*, $-CH(CH_3)CH_3$ vs. $-C(OH)(CH_3)CH_3$). Despite this extremely close structural relationship, as well as the fact that the proposed lead compounds had "remarkable and unexpected potency as antihypertensives" and no indicated deleterious side effects, the court rejected all six compounds as lead compounds. This is because *other* second generation ARBs in the prior art "demonstrated greater potency and all had been more thoroughly studied than the '902 ARBs." The court of *Daiichi* specifically cautioned against the use of hindsight and the reliance on structural similarity alone when selecting a lead compound:

> [T]he attribution of a compound as a lead compound after the fact must avoid hindsight bias; it must look at the state of the art *at the time the invention was made* to find a motivation to select and then modify a lead compound to arrive at the claimed invention [citations omitted]. Accordingly, proving a reason to select a compound as a lead compound depends on more than just structural similarity, but also knowledge in the art of the functional properties and limitations of the prior art compounds. [citation omitted]. ***Potent and promising activity in the prior art trumps mere structural relationships.***

Daiichi at 1354. (Emphasis added.)

In another case, *Otsuka*, the "unsubstituted butoxy" derivative was considered by the Federal Circuit to be of "middling potency" because it was shown to be only the fifth most active compound of nine tested in an assay indicative of potential antipsychotic activity. The Federal

Circuit stressed the importance of a prior art compound's disclosed properties in a lead compound analysis and agreed with the District Court that "[g]enerally, a skilled artisan would be attracted to the most potent compounds in selecting a lead compound for development" and thus disqualified the "unsubstituted butoxy" as a lead compound. *Otsuka* at 1294.

In cases where a structurally close prior art compound did pass the first prong of the lead compound obviousness analysis, the relative activity of that compound was an especially important factor. In *Altana Pharma AG v Teva Pharms. USA, Inc*, 566 F.3d 999, (Fed. Cir. 2009), the court found that compound 12 from the prior art '518 patent was a valid lead compound because it was "one of the more *potent* of the eighteen compounds of the '518 patent for which data was provided during prosecution." *Altana* at 1008 (Emphasis added.). In fact, the district court found and the CAFC agreed "that one of skill in the art would have used the more potent compounds of the '518 patent, including compound 12, as a starting point from which to pursue further development efforts." *Altana* at 1009 (Emphasis added.).

In *Eisai*, the Federal Circuit found that the prior art compound lansoprazole was a suitable lead compound based not only on its structural similarity, but also because it was "twenty times superior to [another prior art compound] omeprazole for anti-ulcer action" and had desirable lipophilicity and low molecular weight. *Eisai* at 1358. Lansoprazole was ultimately found to fail the second part of the analysis because there was no discernible reason to modify it to produce the patented compound rabeprazole. *Id.*

As discussed in more detail below, neither of the starting compounds relied upon by the Examiner, namely the compound of Example 5 of Backer, nor the compound of Example 62 of Sum, satisfies the two-part test of *Takeda.*

III. Application of the Law of Chemical Obviousness to the Cited References

a. Backer

The Examiner has not made out a *prima facie* case of obviousness under the well-established two-part test of *Takeda.* If it *were* true that a person of ordinary skill in the art would have a reasonable expectation that **any** prior art tetracycline would be an effective antibacterial agent, then *no reason* would exist to *specifically* select Compound 5 of Backer out of all active tetracyclines, and *no reason* exists to further modify it, especially by *specifically* replacing the

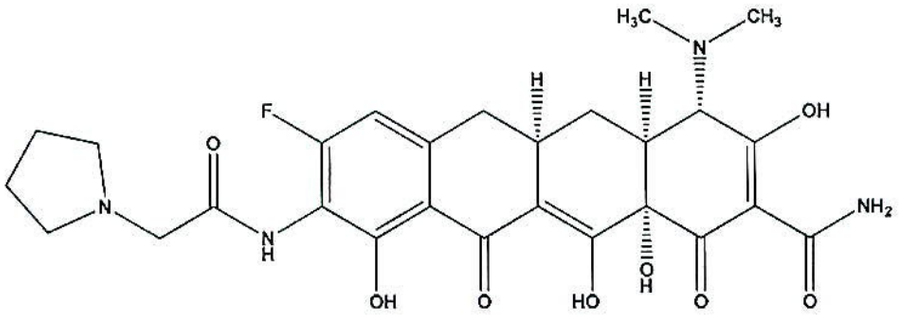
C7 substituent with fluorine. In other words, if **any** tetracycline is expected to have antibacterial activity, then, absent some *specific* direction toward Compound 5 of Backer, *the only reason* to select Compound 5 of Backer and to modify its C7 substituent is knowledge of the structure that would result from such a selection and modification, i.e., TP434. As explained above, it is improper to use Applicants' disclosure to either identify or provide motivation to modify a lead compound.

When the disclosed activity of the compound of Example 5 of Backer is taken into consideration in the first part of the two-part *Takeda* test, the law requires that it be disqualified as a lead compound. Compound 5 is the worst performing antiproliferative of the five compounds tested in each and every one of the three ovarian tumor cell lines and two colon tumor cell lines set forth in Table 2 of Backer. Therefore, like the "unsubstituted butoxy" of *Otsuka* that was rejected as a lead compound because it was of "middling potency," ranking only fifth out of nine, so too should Compound 5 be rejected because it ranked last when compared to all compounds tested.

Moreover, Backer inherently expresses a preference for compounds with the greatest potency, because smaller doses can be used and greater efficacy can be achieved in keeping tumor growth in check after chemotherapy or radiation therapy, thereby requiring fewer treatments. (*See* Backer, Col. 17, lines 54-61.) Therefore, a chemist of ordinary skill in the art would not select the compound of Example 5 of Backer for further development efforts because it would not be the most promising compound to modify in order to improve upon its activity and obtain a compound with better activity.

b. Sum in View of Bandarage

The Examiner rejected Applicants' claimed method over Sum in view of Bandarage by comparing TP-434 to a hypothetical tetracycline that differs from the compound of Example 62 of Sum by substitution of the C8 chloro group of Example 62 with a fluoro group extracted from the generic definition of the variable X of Formula I of Sum. The Examiner then asserted that Bandarage teaches equivalence in the position of a halogen at C7 and C8 of a tetracycline compound and that, therefore TP-434 is obvious in view of the hypothetical compound derived from Example 62.



Hypothetical compound

The hypothetical compound proposed by the Examiner is *not a prior art compound, because it is not found anywhere in the art of record.* In addition, there is no exemplification in Sum of compounds having *any* halo group at C7. Further, Sum does not motivate a skilled artisan to remove its mandatory substituent at C8, and, likewise, Bandarage has no specific teaching directing a skilled artisan toward such a modification. Moreover, combining the teachings of Sum and Bandarage would require removing Sum's mandatory substituent at C8.

In particular, there is no motivation provided in Sum to modify the compound of Example 62 in a manner required to arrive at TP-434 (*i.e.*, to place a fluoro substituent at C7, and to remove a substituent at C8). Rather, Sum describes a chemical genus that *requires* a C8 substituent in a position that is unsubstituted in TP-434. Within the chemical genus of Sum, the C7 substituent includes a generically defined substituent R (*see* Formula (I) of Sum). Even though a "halogen" is a permitted value for substituent R at C7 of Sum, there is a clear preference in Sum for halogen atoms *other than* fluoro. Specifically, at page 6, lines 27-30 of Sum, fluorine is omitted from the list of preferred values of the C7 substituent. There also is *no exemplification* in Sum of compounds having a halo group at C7; the compounds specifically exemplified in Sum only possess $-NMe_2$ or -H at C7.

Bandarage is relied upon by the Examiner for its teaching of possible values for its C7 and C8 substituents. However, as Applicants previously argued, Bandarage recites a broad Markush group for each of the C7 and C8 substituents, which among the many choices include hydrogen and halogen. The Markush group that defines substituent R^9 of Bandarage at the C9 position of the tetracycline core is co-extensive with the Markush groups that define the C7 and C8 substituents (see Bandarage, col. 4, ll. 31-35). There is no specific teaching or motivation in

Bandarage, or any other reference cited by the Examiner, to select positions C7 and C8 out of the three positions (C7, C8, and C9) for modifying their respective substituents, or to select hydrogen and halogens, out of all possible values for the three substituents at C7, C8, and C9, as possible modifying values. There also is no disclosure or suggestion, nor is there any motivation to select the *specific* position of hydrogen and a fluoro group to obtain the compound of Applicants' claimed method, nor its positional isomer, as hypothesized by the Examiner.

Thus, Bandarage also does not provide the specific teaching necessary to motivate one of ordinary skill in the art to modify the compound of Example 62 of Sum in a manner necessary to obtain TP-434, nor is there any teaching or motivation to modify the hypothetical compound the Examiner derived from Example 62. As stated in In re Geiger, 815 F.2d 686 (Fed. Cir.):

> It is not required that the claimed invention be compared with subject matter that does not exist in the prior art. The applicant is not required to create prior art, nor to prove that his invention would have been obvious if the prior art were different than it actually was.

In re Geiger at 689 (Newman, J., concurring.)

As recently emphasized by the Federal Circuit in the *Pfizer, Inc. v., TEVA Pharm. USA, Inc.*, cited above, the second prong of the *Takeda* test is whether the prior art would have taught "'***specific molecular modifications***' to a lead compound." None of the cited references taken separately or in combination with each other provides the requisite *specific* teachings.

c. Backer, Sum and Bandarage in Combination

Combining Backer, Sum and Bandarage fails to overcome the shortcomings of the prior two rejections discussed above. In particular, there is nothing in Sum or Bandarage that teaches or suggests that any of the Backer compounds would have antibacterial activity. The weakly anti-angiogenic Example 5 compound of Backer relied upon by the Examiner as the starting compound to modify to obtain the compound recited in present Claim 1 lacks a C8 substituent. In contrast, Sum requires a C8 substituent for its antibacterial compounds, while Bandarage fails

to provide any antibacterial data for its compounds and therefore suggests nothing about the activity of the Backer compounds.

Conversely, Backer does not remedy the deficiencies of Sum and Bandarage, taken separately or in any combination. In particular, nothing in Backer suggests the equivalence between hydrogen and halogen at the C7 and C8 positions in general or with respect to antibacterial activity, on which Backer is silent. All of Backer's compounds *require* a substituent at C7 and are unsubstituted at C8.

In summary, Sum and Bandarage fail to disclose or suggest (1) selecting the compound of Example 5 of Backer or (2) the specific modifications to the compound of Example 5 required to arrive at TP-434. Backer fails to disclose or suggest (1) selecting the compound of Example 62 of Sum or (2) the specific modifications to the compound of Example 62 required to arrive at TP-434. Nor has the Examiner provided the basis for any motivation to make the specific molecular modification as required by the court in *Takeda*.

In view of the foregoing, the cited references, taken separately or in any combination, fail to establish a *prima facie* case of obviousness of the subject matter of Applicants' pending claims. Withdrawal of the rejections under 35 U.S.C. §103(a) are requested.

IV. Summary of Reply to Obviousness Rejection Under 35 U.S.C. §103(a)

The Examiner has not established a *prima facie* case of obviousness of Applicants' claimed method over the cited references. Even if a *prima facie* of obviousness were established in view of the closest prior art compound, namely the compound of Example 5 of Backer, Applicants have rebutted that *prima facie* case by presentation of evidence demonstrating an appreciable difference and substantially improved properties over the compound of Example 5 of Backer being relied upon by the Examiner to make the *prima facie* case. Under controlling law, the Examiner *must* consider the evidence presented by Applicants' in the First and Second Sutcliffe Declarations. Moreover, absent evidence to the contrary, which the Examiner has not presented, Applicants' demonstrated substantially improved properties are *ipso facto* unexpected, thereby rebutting any *prima facie* case of obviousness as a matter of law. Therefore, Applicants' Claim 1 and claims dependent thereon meet the requirement of 35 U.S.C. §103(a), and Applicants respectfully request reconsideration and withdrawal of this rejection of these claims.

CONCLUSION

In view of the above amendments and remarks, it is believed that all claims are in condition for allowance, and it is respectfully requested that the application be passed to issue. If the Examiner feels that a telephone conference would expedite prosecution of this case, the Examiner is invited to call the undersigned.

Respectfully submitted,
HAMILTON, BROOK, SMITH & REYNOLDS, P.C.

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Concord, MA 01742-9133
Date: March 21, 2014

SMA
August 23, 2013

PATENT APPLICATION
Attorney's Docket No.: 4142.1017-006

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Inventors:	Jingye Zhou, Xiao-Yi Xiao, Louis Plamondon, Diana Katharine Hunt, Roger B. Clark and Robert Zahler		
Application No.:	12/462,795	Group:	1628
Filed:	August 7, 2009	Examiner:	Badio, Barbara P.
Confirmation No:	5985		
For:	C7-Fluoro Substituted Tetracycline Compounds		

CERTIFICATE OF MAILING OR TRANSMISSION

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Typed or printed name of person signing certificate

DECLARATION OF JOYCE A. SUTCLIFFE, Ph.D. UNDER 37 CFR § 1.132

Mail Stop RCE
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

The undersigned, Joyce A. Sutcliffe, Ph.D., hereby declares that:

1. I am the Senior Vice President of Biology at Tetraphase Pharmaceuticals, Inc. (hereinafter "Tetraphase"), the assignee of the above-noted application. I have worked at Tetraphase since May 2009. I received my Ph.D. in microbiology from the University of Florida, Gainesville. After obtaining my degree, I held postdoctoral positions at the University of Massachusetts

EXHIBIT A
13/718,909

Medical School and the National Institutes of Health. Prior to my employment by Tetraphase, I was Vice President, Research, at NanoBio Corporation, where I led antimicrobial, antiviral and antifungal discovery activities. In addition, I was Chief Research Scientist and Vice President, Biology, at Rib-X Pharmaceuticals from 2001 to 2007 and spent sixteen years at Pfizer conducting research in the area of antibiotic discovery and development.

2. My work at Tetraphase focuses on novel antibiotics effective against serious and life-threatening infections such as multidrug-resistant (MDR) infections. Developing such antibiotics is a pressing and unmet medical need, particularly for MDR Gram-negative bacteria (*see* Appendix 1). One such antibiotic I work on at Tetraphase is commonly referred to as TP-434 or eravacycline. The composition of matter for TP-434 and pharmaceutical compositions comprising TP-434 are the subject matter claimed in the above-noted application (the "Application").

3. TP-434 has broad spectrum antibacterial activity and has recently demonstrated a high clinical cure rate[1] against a wide variety of multidrug-resistant Gram-negative and Gram-positive aerobic and anaerobic bacteria in a Phase 2 human clinical trial (IV administration) for the treatment of patients with complicated intra-abdominal infections (cIAI) (*see* Appendix 2 for results). In addition, the FDA has designated TP-434 as a Qualified Infectious Disease Product (QIDP)[2] and the Infectious Diseases Society of America (IDSA) has identified TP-434 as one of only seven new drugs in development for the treatment of infections caused by Gram-negative bacteria (*see* Appendix 3).

4. As indicated in Paragraph 2 above, I am familiar with the Application and I am aware that in a Final Office Action issued from the U.S. Patent Office on July 24, 2012 (the "Final Office Action"), the Examiner rejected the claims as allegedly obvious under 35 U.S.C. §103(a) over Backer *et al.* (U.S. Patent No. 5,574,026). From my review of the Office Action, I understand the Examiner's rejection to be that the claimed compound (TP-434) and Example 5 of U.S. Patent No. 5,574,026 (the "Backer Compound") are structurally similar compounds and would

[1] Cure rate was based on the Microbiologically Evaluable (ME) population.

[2] QIDP designation was granted for treatment of complicated intra-abdominal infections (cIAI) and complicated urinary tract infections via IV administration.

be expected to have similar activity. Structural representations of TP-434 and the Backer Compound are presented in Appendix 4.

5. I make this declaration to present the results of *in vitro* testing performed at Tetraphase under my direction to determine the Minimum Inhibitory Concentration (MIC) values[3] for TP-434 and the Backer Compound against a variety of bacterial strains. I also make this declaration to show, based upon those results, that TP-434 has unexpectedly superior antibacterial activity in comparison to the Backer Compound.

6. The results presented in Appendix 5 were generated according to the Clinical and Laboratory Standards Institute (CLSI) performance standards using the MIC assay and bacterial strains set forth in Example 13 of the above-noted Application. The MIC values correlate with antibacterial activity, with a lower value being indicative of greater potency. For completeness, Appendix 6 describes the details of the strains used to generate the results reported herein, as well as an abbreviated description of the MIC assay used to compare the two compounds.

7. The Backer Compound was prepared following the procedures in U.S. Patent No. 5,574,026 to Backer *et al.* Details of the synthesis and characterization of the Backer Compound can be found in Appendix 7. TP-434 was prepared according to the procedure set forth in the specification of the above-noted application.

8. Appendix 5 compares the results of testing TP-434 and the Backer Compound against a diverse panel of bacteria that included both Gram-positive and Gram-negative bacteria. The panel of bacteria is the same as the panel used in Example 13 of the above-noted application. The shaded areas of Appendix 5 point to each instance where TP-434 is unexpectedly more potent than the Backer Compound. The bottom row of the table shown in Appendix 5 provides the fold increase in potency of TP-434 over the Backer Compound.

9. The results in Appendix 5 show that TP-434 demonstrates superior potency in comparison to the Backer compound in 12 of the 15 bacterial strains tested (see shaded areas). The fold

[3] Minimum Inhibitory Concentration (MIC) values are defined as the lowest concentration of an antimicrobial that will inhibit the visible growth of a microorganism after overnight incubation (18-24 hours at 35°C±2°C). MICs are used to determine the *in vitro* activity of antimicrobials with lower MICs values indicative of greater potency.

increase in potency corresponding to the 12 shaded areas in Appendix 5 range from a value of 2 to a value of 8, with the majority of the values indicating a 4 fold or more increase.[4] It is important to note that in all of the tetracycline resistant strains where a conclusion of relative potency can be made (strains indicated by an asterisk in Appendix 5), TP-434 is 4 to 8 fold more potent than the Backer Compound.

10. MIC values for both compounds against two of the remaining strains (SP106 and SP160) are at the lower numerical limit of the assay and no conclusion with respect to relative potency of TP-434 and the Backer Compound can be made. In the one remaining strain, TP-434 and the Backer Compound show equivalent potency.

11. Based on the results presented in Appendix 5 and discussed in Paragraph 9 above, it is my opinion that the antibacterial potency of TP-434 is unexpectedly superior when compared to the Backer Compound. This is because I would not have expected the replacement of the iodo substituent in the Backer Compound with the fluoro substituent of TP-434 to alter potency. The fact that TP-434 shows superior potency in 80% (12 of 15) of the bacterial strains tested and, to the extent discernible in the present assay, equivalence in the other three strains, is an unexpected result.

12. I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: 8/23/2013

Joyce A. Sutcliffe

Joyce A. Sutcliffe, Ph.D.

[4] In strains where the MIC of TP-434 was at or below the lowest numerical limit of the assay and the MIC of the Backer Compound was above this limit, the fold increase in indicated as "2 or more"; where the MIC of the Backer Compound was above the highest numerical limit of the assay and the MIC of TP-434 was below this limit, the fold increase is indicated as "more than 2."

APPENDIX 1: Hersh AL, Newland JG, Beekmann SE, Polgreen PM, Gilbert DN. 2012. Unmet medical need in infectious diseases. Clin Infect Dis 54:1677-1678.

APPENDIX 2: Poster L1-1647a, Presented at the 52nd Annual Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC), September 9-12, 2012, J. Solomkin, G. Cesnauskas, M. Ramesh, S. Walpole, J. Sutcliffe, P. Horn, "Efficacy and Safety of TP-434 (Eravacycline) versus Ertapenem in Complicated Intra-Abdominal Infection (cIAI)"

APPENDIX 3: Clinical Infectious Diseases (Advance Access, published April 17, 2013), Helen W. Boucher *et al.*, 10 x '20 Progress-Development of New Drugs Active Against Gram-Negative Bacilli: An Update From the Infectious Diseases Society of America"

APPENDIX 4: Structural Representations of TP-434 and Backer Compound

APPENDIX 5: Results of testing TP-434 and Backer Compound in MIC Assay.

APPENDIX 6: MIC Assay and Bacterial Strains

APPENDIX 7: Description of Synthesis used to Prepare Backer Compound

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PLANILLA DE RADICACIÓN - DIGITALIZACIÓN DIARIA
RELACIÓN DE ANEXOS

Dependencia:	Fecha: ____/____/____ AAAA MM DD	Recorrido:
2020	2014 7 11	

Tipo de Radicación: Entrada: ☐ Salida: ☐ Traslado: ☐ Convenio: ☐

Radicador: | Planilla:

ID	No. Radicación	Consecutivo	Item
1	11 - 27711 -	20	
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			
17			

Item	Unidad de Conservación
1	Bolsa
2	Caja
3	Libro
4	Sobre
5	Tomo
6	Otro
Item	*Soporte Documental*
7	CD
8	Disquete
9	DVD
10	Folleto
11	Foto
12	Periódico
13	Plano
14	Publicación Periódica
15	USB
16	Otro

Observaciones:

Folio 77 CD