

SEÑOR

SUPERINTENDENTE DE INDUSTRIA Y COMERCIO

E. S. D.

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REF: Expediente 00-065-489

TÍTULO SOLICITADO: "Una composición farmacéutica para ser usada como anticonceptivo"

SOLICITANTE: Schering Aktiengesellschaft

APODERADO: Margarita Castellanos Abondano

TRÁMITE: Remisión de información

JOSÉ LUIS REYES VILLAMIZAR, mayor de edad, identificado como aparece al pie de la firma, abogado en ejercicio, titular de la tarjeta profesional número 44655 del C.S.J, obrando en nombre y representación de **LABORATORIO FRANCO COLOMBIANO S.A. – LAFRANCOL S.A.**, sociedad comercial constituida y vigente de acuerdo con las leyes de la República de Colombia, con domicilio y sede principal en Cali, hechos que constan en el poder y en el certificado de existencia y representación legal anexos, de manera respetuosa, y con el propósito de coadyuvar en la decisión que sobre la solicitud de patente de la referencia deberá adoptarse próximamente, por este escrito me permito remitir información fundamentada que demuestra que la solicitud en mención no cumple los requisitos de novedad y nivel inventivo exigidos por la Decisión 486 de 2000, y que por ello debería el Despacho abstenerse de concederle el privilegio de patente solicitado.

Además de la defensa del orden jurídico vigente en materia de propiedad industrial, el presente escrito se fundamenta en la necesidad de evitar que la concesión de la solicitud vulnere la libre competencia y restrinja el mercado farmacéutico, tanto en perjuicio de los legítimos intereses comerciales de **LAFRANCOL**, como de los consumidores colombianos en general.

Para el efecto me permito hacer una breve descripción de antecedentes relacionados con la materia que se reivindica en la solicitud de la referencia, con el

fin de demostrar la carencia absoluta de novedad y de nivel inventivo de la misma.
Veamos:

1.1. La solicitud 00-065-489 carece de novedad y de nivel inventivo por la materia revelada en la publicación PCT WO 95/07081 de 16/03/1995:

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 95/07081 presentada 8/09/1994 y publicada 16/03/1995
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, Drospirenona (<i>un progestágeno, anotamos</i>), en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox. 2mg hasta 4mg, y, como segundo ingrediente activo, etinilestradiol (<i>un estrógeno, anotamos</i>) en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de Drospirenona correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y etinilestradiol en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	Rv. 1: Preparación oral para terapia de sustitución y para contracepción que comprende al menos un progestágeno y al menos un estrógeno en donde la dosis del estrógeno varia con una periodicidad tal que se evita substancialmente la pérdida de sangre. Rv. 6: Preparación donde el progestágeno comprende: Progesterona 300-900mg/día, Noretisterona acetato 0,2-5mg/día, Medroxiprogesterona acetato 1-5mg/día, d-norgestrel 30-150mg/día, desogestrel 30-150mg/día, norgestimato 30-150mg/día, ciproterona acetato 0,2-2mg/día, gestodeno 10-150mg/día, 3-cetodesogestrel 10-150mg/día, Drospirenona 0,2-3.0mg/día, o combinación de los mismos. Rv. 7: Preparación donde el estrógeno comprende: Etinilestradiol 5-15 gamma/día¹, valerato de estradiol 1-4mg/día; estradiol 1-2mg/día; estrógenos conjugados 0,3-1.25mg/día; estriol 1-4mg/día; o combinación de los mismos.

Una sencilla comparación entre los documentos citados lleva a concluir que la composición revelada en la solicitud en trámite, que comprende Drospirenona y Etinilestradiol carece por completo de novedad y nivel inventivo, puesto que la

¹ 1 Gamma= 1 µg. (www.smfc-fercofiv.ma/pilules.htm)

mencionada publicación PCT WO 95/07081 ya revelaba la misma composición conformada por un progestágeno como Drospirenona y un estrógeno como Etinilestradiol en rangos similares a los que se pretende patentar en la solicitud que nos ocupa, pues en la primera la Drospirenona está presente en un rango comprendido entre 0.2-3.0 mg/día, al paso que en la solicitud en trámite una dosis preferida del mismo principio activo es de 3.0mg, evidentemente comprendido en el rango anterior.

Respecto de las dosis mencionadas, hago notar que la eventual concesión de la solicitud con base en las cantidades de los compuestos "Etinilestradiol" y "Drospirenona" empleados en la formulación, además de versar sobre un tratamiento terapéutico basado en la administración simultánea de dos principios activos para tratar una patología determinada –asunto expresamente prohibido por la norma andina-, supondría restringir injustificadamente a los médicos y desarrolladores de medicamentos la posibilidad de proponer y utilizar formulaciones de **etinilestradiol** con una dosis diaria en el rango desde aproximadamente 0.01mg hasta 0.05mg y de **Drospirenona** con una dosis diaria en el rango desde 2mg hasta 4mg.

1.2. La solicitud 00-065-489 carece de novedad y de nivel inventivo por la materia revelada en la publicación US 5,756,490 de 26/05/1998, equivalente a PCT WO 95/26730 presentada en 30/03/1995

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	US 5,756,490 publicada 26/05/1998 equivalente a PCT WO 95/26730 presentada 30/03/1995
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g.	Rv. 1: Una preparación farmacéutica combinada con dos componentes (...) y que esto es querido durante el tiempo la administración secuencial oral, que en cada caso consiste en un número de unidades de dosificación diarias que son colocadas físicamente separadamente e individualmente en una unidad de embalaje desprendible, por el cual como primer componente hormonal contiene la combinación de una preparación de estrógeno en al menos una dosificación que es suficiente para inhibir la ovulación y una preparación de un gestageno

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	US 5,756,490 publicada 26/05/1998 equivalente a PCT WO 95/26730 presentada 30/03/1995
Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	en estructura de una fase o en múltiple fase y como un segundo componente hormonal contiene únicamente una preparación estrogénica, donde el primer componente hormonal comprende 23 o 24 unidades de dosis y el segundo componente hormonal comprende 4 a 10 unidades diarias, las unidades diarias del primer componente hormonal no contiene la combinación de un estrógeno biogénico y un estrógeno sintético, y el número total de unidades diarias es igual a el número total de días de el ciclo deseado, por al menos 28 días en longitud. Rv. 2: Preparación combinada de acuerdo con la Rv. 1, donde el estrógeno de el primer componente hormonal es seleccionado del grupo de: 17β estradiol, etinilestradiol y 17β-estradiol valerato y el gestágeno es seleccionado del grupo de compuestos: gestodeno, levonogestrel, desogestrel, 3-ketodesogestrel, <u>Drospirenona</u> (...) y el estrógeno del segundo componente hormonal es seleccionado del grupo de compuestos: 17β estradiol, <u>etinilestradiol</u> y 17β-estradiol valerato. Rv. 3: Preparación combinada de acuerdo con la Rv. 2 donde el estrógeno del primer componente hormonal en cada unidad de dosis diaria contiene una dosis de: 1.0 a 6.0mg 17β estradiol, 0.015 a 0.025mg etinilestradiol y 1.0 a 4.0mg 17β-estradiol valerato y el gestágeno en cada unidad de dosis diaria contiene una dosis de: 0.05 a 0.075mg gestodeno, 0.05 a 0.125mg levonogestrel, 0.06

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	a 0.15mg desogestrel, 0.06 a 0.15mg 3-ketodesogestrel, <u>1.0 a 3.0mg Drospirenona</u>.... Rv. 4 y 5: La preparación combinada..., donde el segundo componente hormonal contiene, en cada unidad de dosis, una cantidad de: 1.0 a 6.0mg 17β estradiol, <u>0.002 a 0.04mg etinilestradiol</u> y 1.0 a 4.0mg 17β-estradiol valerato. Rv. 6: Preparación combinada donde el segundo componente hormonal contiene <u>etinilestradiol</u> en una cantidad de <u>0.01 a 0.025mg</u>
Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación colocadas en una unidad de empaque, empaquetadas separadamente y separables individualmente y provistas para la administración oral durante <u>un periodo de 21 días consecutivos</u>..... Rv. 11: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación diaria colocadas en una unidad de empaque, empaquetadas separadamente y separables individualmente, y provistas para la administración oral durante <u>un periodo de 21 días consecutivos</u> Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al período de al menos 21 días consecutivos, siendo el número	Rv. 1: (ver más arriba) Rv. 8: Preparación combinada de acuerdo con la Rv. 1, donde el número total de <u>unidades diarias de hormona es 28</u>. Rv. 9: La preparación combinada de acuerdo con la Rv. 1, donde el primer componente hormonal <u>tiene 24 unidades diarias y el segundo componente hormonal tiene 4-8 unidades diarias</u>. Rv. 10: La preparación combinada de acuerdo con la Rv. 1, donde el primer componente hormonal <u>tiene 24 unidades diarias y el segundo componente hormonal tiene 4 unidades diarias</u>.

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total de unidades de dosificación diaria <u>al menos 28</u> Rv. 16: La preparación de acuerdo con cualquiera de las Rv.s 10 a 14, caracterizada porque el número de unidades de dosificación diaria que comprenden la combinación de Drospirenona y Etinilestradiol es 21, 22, 23 o 24, y porque el número de unidades de dosificación diaria que no contienen agente activo es 7, 6, 5 ó 4. Rvs. 34, 35, 36, 38, 39	

No hacen falta mayores análisis para concluir que la patente US 5,756,490, equivalente a la solicitud WO 95/26730 ya revelaba la existencia de una composición farmacéutica formada por Etinilestradiol y Drospirenona, tal como se reivindica en la solicitud de patente que nos ocupa, asunto que aniquila nuevamente la novedad y el nivel inventivo de la misma.

Del mismo modo, se llama la atención al hecho de que los rangos de dosificación propuestos en la solicitud para dichos compuestos "Etinilestradiol" y "Drospirenona" y el periodo de administración de los mismos, ya habían sido revelados en la patente US 5,756,490, pues en ésta se emplean formulaciones de etinilestradiol con una dosis diaria en el rango de 0.010 a 0.025mg y de Drospirenona con una dosis diaria en el rango de 1.0 a 3mg y se administran en unidades diarias de 28 ó de 24 para el primer componente hormonal y 4 a 8 para el segundo componente hormonal. También reitero que las dosis en cuestión versan sobre un método de tratamiento de prohibido patentamiento según la norma andina-, y hago notar que aún si tales métodos fuesen patentables, en este caso carecerían de Nivel Inventivo, pues la definición de rangos de dosis y la determinación del período más adecuado para su administración son pasos necesarios y previos a la formulación, y resultan obvios para una persona con medianos conocimientos técnicos en la materia.

1.3. La solicitud 00-065-489 carece de novedad y de nivel inventivo por la materia revelada en el documento PCT WO 98/04267, publicado en 5/02/1998

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 98/04267 presentada 23/07/1997 y publicada 5/02/1998
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	Rv. 1: Un método de contracepción de una progestina en una dosis diaria seleccionada del grupo que consiste de 40-500µg trimegestone, 250µg- 4µg dienogest, y 250µg-4mg Drospirenona, y un estrógeno en una dosis diaria equivalente en actividad estrogénica a 10-20µg de etinilestradiol para 23-25-días empezando en el día 1 del ciclo menstrual; en el que la misma dosificación de la combinación de progestina y estrógeno es administrado en cada uno de los 23-25 días, seguida por la administración de un estrógeno en una dosis diaria equivalente en actividad estrogénica a 5-15µg de etinilestradiol por 3-5 días; en el que la misma dosis de el estrógeno es administrada encada uno de los 3-5 días; tal que el número de días de administración de la combinación de progestina y estrógeno más el número de días de administración del estrógeno es igual a 28 por ciclo menstrual. Rv. 3: El método de acuerdo con la Rv. 2, donde el estrógeno es Etinilestradiol Rv. 5: El método de acuerdo con la Rv. 4, donde la dosis diaria de Etinilestradiol durante el período de 24 días de administración es 15µg. Rv. 18: El método de acuerdo con la Rv. 5, donde la progestina es Drospirenona. Rv.19, 20, 21 y 22: El método de acuerdo con la Rv. 18, donde la dosis diaria de Drospirenona es 3mg, 2mg, 1mg y 500µg.

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Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación.... Y provistas para la administración oral durante <u>un período de 21 días consecutivos</u>..... Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al periodo de al menos 21 días consecutivos, siendo el número total de unidades de dosificación diaria <u>al menos 28</u>	Rv. 1: (ver más arriba) Rv. 4: El método de acuerdo con la Rv. 3, donde la combinación progestina/estrógeno es administrada por 24 días por ciclo menstrual empezando sobre el primer día del ciclo menstrual. Rv. 23: Un kit contraceptivo adaptado para administración oral diaria, la cual comprende 28 unidades de dosificación separadas, 23-25 unidades de dosis separadas, dichas unidades de dosis cada una contiene una combinación de progestina en una dosis seleccionada del grupo que consiste de de 40-500µg trimegestone, 250µg- 4µg dienogest, y 250µg-4mg Drospirenona, y un estrógeno en una dosis diaria equivalente en actividad estrogénica a 10-20µg de etinilestradiol, donde cada unidad de dosificación contiene la misma dosificación de progestina y estrógeno; y 3-5 días de dichas unidades de dosificación contienen un estrógeno en una dosis diaria equivalente en actividad estrogénica a 5-15µg de etinilestradiol, en el que cada 3-5 días de unidades de dosificación contienen la misma dosificación de estrógeno. Rv. 26: El kit contraceptivo de acuerdo con la Rv. 25, donde 24 de las 28 unidades de dosificación contienen una combinación de progestina/estrógeno, y 4 de las unidades de dosificación están libres de progestina.

Lo anterior demuestra que la solicitud PCT WO 98/04267, ya revelaba la existencia de una composición farmacéutica formada por un estrógeno como Etinilestradiol y una progestina como Drospirenona, tal como lo quiere hacer el solicitante mediante la actual solicitud de patente colombiana; en

consecuencia éste último documento carece por completo de novedad y de nivel inventivo.

Del mismo modo, los rangos de dosificación propuestos para dichos compuestos "Etinilestradiol" y "Drospirenona" y el periodo de administración de los mismos, ya habían sido revelados en la PCT WO 98/04267, en la cual se utilizan formulaciones de **etinilestradiol** con una dosis diaria en el rango de 10-20µg (0.01mg-0.02mg) y de **Drospirenona** con una dosis diaria en el rango de 250µg-4mg (0.25mg-4mg) y se administran entre 23 y 25 días del ciclo menstrual.

1.4. La solicitud 00-065-489 carece de novedad y de nivel inventivo por la materia revelada en el documento PCT WO 98/27929, publicado en 02/07/1998

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 98/27929 presentada 22/12/1997 y publicada el 2/07/1998
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a	Rv. 1: Empleo de gestágenos terapéuticos en la producción de un medicamento para el tratamiento de los desórdenes disfóricos premenstruales (PMDD). Rv. 2: Empleo de <u>Drospirenona</u>, Ciproteronacetato, Dienogest de acuerdo con la Reivindicación 1. Rv. 3: Empleo de acuerdo con la Reivindicación 1, junto con un <u>Estrógeno</u>. Rv. 5: Empleo de acuerdo con la Reivindicación 3, junto con <u>Etinilestradiol</u>. Rv. 11: Empleo de <u>Drospirenona</u> de acuerdo con la Reivindicación 1, en una cantidad de <u>0,5mg y menos de 5mg</u> diarios. Rv. 12: Empleo de <u>Etinilestradiol</u> de acuerdo con la Reivindicación 5, en una cantidad de <u>0,010mg y 0,05mg</u> diarios.

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una dosis diaria de 0.03mg.	
Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación.... Y provistas para la administración oral durante <u>un período de 21 días consecutivos</u>..... Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al período de al menos 21 días consecutivos, siendo el número total de unidades de dosificación diaria <u>al menos 28</u>	Rv. 10: Empleo según la Reivindicación 9 entre <u>10 y 28</u> de ciclo menstrual. (Rv. 9: Empleo según la Reivindicación 1, exclusivamente durante la fase luteana del ciclo menstrual)

Lo anterior demuestra que la solicitud PCT WO 98/27929, ya revelaba la existencia de una composición farmacéutica formada por un estrógeno como Etinilestradiol y un gestágeno como Drospirenona, tal como lo quiere hacer el solicitante mediante la actual solicitud de patente colombiana; en consecuencia éste último documento carece por completo de novedad y de nivel inventivo.

Del mismo modo, los rangos de dosificación propuestos para dichos compuestos "Etinilestradiol" y "Drospirenona" y el periodo de administración de los mismos, ya habían sido revelados en la PCT WO 98/27929, en la cual se utilizan formulaciones de etinilestradiol con una dosis diaria en el rango de 0.010 a 0.05mg y de Drospirenona con una dosis diaria en el rango de 0.5 a 5mg y se administran entre 10 y 28 días del ciclo menstrual.

1.5. La solicitud 00-065-489 carece de novedad y nivel inventivo por la materia revelada en el documento US 5,583,129, publicado en 10/12/1996

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	US 5,583,129 publicada 10/12/1996, equivalente a PCT WO 95/17194 presentada 22/12/1994 y publicada el 29/06/1995
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	Rv. 1: Un método para inducir la contracepción en una mujer en edad reproductiva quien aún no ha alcanzado la premenopausia, que comprende la administración de una composición que comprende un estrógeno seleccionado de: 2.0 a 6.0mg de 17β-estradiol y 0.015 a 0.020mg de <u>etinilestradiol</u>; y un gestágeno seleccionado de: 0.05 a 0.075mg de gestodeno; 0.075 a 0.125mg de levonorgestrel, 0.06 a 0.15mg de desogestrel; 0.06 a 0.15mg de 3-ketodesogestrel; 0.1 a 0.3mg de <u>Drospirenona</u>; 0,1 a 0,2mg de acetato de ciproterona; 0,2 a 0,3mg de norgestimate y >0.35 a 0.75mg de noretisterona, donde la composición es administrada por 23 o 24 días, empezando sobre el primer día del ciclo menstrual, seguido por 5 o 4 píldoras libre o píldora de azúcar, durante in total de 28 días en el ciclo de administración. Rv. 2: Un método de acuerdo con la Rv.1, donde el estrógeno es <u>Etinilestradiol</u>. Rv. 6: Un método de acuerdo con la Rv.1, donde el gestágeno es Ciproterona o <u>Drospirenona</u>. Rv. 8: Un método de acuerdo con la Rv.1, en donde el estrógeno está presente en una dosis de 20μg de Etinilestradiol o una dosis equivalente de 17β-estradiol y el gestágeno está presente en una dosis de 75μg de gestodeno o una dosis equivalente de levonorgestrel, ciproterona acetato o Drospirenona.

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	US 5,583,129 publicada 10/12/1996, equivalente a PCT WO 95/17194 presentada 22/12/1994 y publicada el 29/06/1995
Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación.... y provistas para la administración oral durante <u>un período de 21 días consecutivos (...)</u> Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al período de al menos 21 días consecutivos, siendo el número total de unidades de dosificación diaria <u>al menos 28</u>	Rv. 1: Un método para inducir la contracepción en una mujer en edad reproductiva quien aún no ha alcanzado la premenopausia, que comprende la administración de una composición que comprende un estrógeno seleccionado de: 2.0 a 6.0mg de 17β-estradiol y 0.015 a 0.020mg de etinilestradiol; y un gestágeno seleccionado de: 0.05 a 0.075mg de gestodeno; 0.075 a 0.125mg de levonorgestrel, 0.06 a 0.15mg de desogestrel; 0.06 a 0.15mg de 3-ketodesogestrel; 0.1 a 0.3mg de Drospirenona; 0,1 a 0,2mg de acetato de ciproterona; 0,2 a 0,3mg de norgestimate y >0.35 a 0.75mg de noretisterona, donde la composición es administrada por <u>23 o 24 días</u>, empezando sobre el primer día del ciclo menstrual, seguido por 5 o 4 píldoras libre o píldora de azúcar, durante un total de <u>28 días</u> en el ciclo de administración.

Una sencilla comparación entre los documentos citados lleva a concluir que la composición revelada en la solicitud en trámite, que comprende Drospirenona y Etinilestradiol carece por completo de novedad y de nivel inventivo puesto que la mencionada patente US 5,583,129 ya revelaba la misma composición conformada por un gestágeno como Drospirenona y un estrógeno como Etinilestradiol en rangos muy cercanos (por lo menos para Etinilestradiol) a los que se pretende patentar en la solicitud que nos ocupa, pues en la primera el Etinilestradiol está presente en un rango comprendido entre 0.015 y 0.020mg, al paso que en la solicitud en trámite una dosis preferida del mismo principio activo es de 0.01mg hasta 0.05mg, preferiblemente 0.15 y 0.03mg, evidentemente comprendido en el rango anterior. Para el caso de Drospirenona, está presente en un rango comprendido entre 0.1 y 0.3mg, valores inferiores a los propuestos en la solicitud en trámite 2.0 a 4.0mg, preferiblemente 3.0 a 3.5mg, pero que en ningún momento desmiente la posibilidad de dichos compuestos activos.

Vale anotar, nuevamente respecto de las dosis, que la eventual concesión de la solicitud con base en los rangos de dosificación de dichos compuestos “Etinilestradiol” y “Drospirenona”, además de versar sobre el uso para el tratamiento terapéutico de una patología determinada -prohibido por la norma andina-, supondría restringir a los médicos y desarrolladores de medicamentos la posibilidad de proponer y utilizar formulaciones de **Etinilestradiol y Drospirenona** en los rangos de dosis propuestos.

1.6. La solicitud 00-065-489 carece de novedad y nivel inventivo por la materia revelada en el documento PCT WO 98/04269,¹ publicado en 05/02/1998:

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 98/04269 presentada 23/07/1997 y publicada el 5/02/1998
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	Rv. 1: Un método de contracepción el cual comprende la administración a una mujer una combinación de una progestina en una dosis diaria seleccionada del grupo que consiste de 40-500µg trimegestona, 250µg-4mg dienogest, y 250µg-4mg <u>Drospirenona</u>, y un estrógeno en una dosis diaria equivalente en actividad estrogénica a 10-20µg <u>Etinilestradiol</u> para 23-25 días empezando sobre el primer día del ciclo menstrual, y donde la misma dosis de la combinación de progestina y el estrógeno es administrada en cada uno de los 23-25 días. Rv. 3: El método de acuerdo con la Rv.2, donde el estrógeno es <u>Etinilestradiol</u>. Rv. 5: El método de acuerdo con la Rv.4, donde la dosis diaria de etinilestradiol es 15µg. Rv. 18: El método de acuerdo con la Rv.5, donde la progestina es <u>Drospirenona</u>. Rv. 19, 20, 21, 22: El método de acuerdo con la Rv.18, donde la dosis diaria de Drospirenona es 3mg, 2mg, 1mg

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 98/04269 presentada 23/07/1997 y publicada el 5/02/1998
	y 500µg.
Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación.... y provistas para la administración oral durante <u>un período de 21 días consecutivos</u> (...). Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al período de al menos 21 días consecutivos, siendo el número total de unidades de dosificación diaria <u>al menos 28</u> Rv. 16: La preparación de acuerdo con cualquiera de las Rv.s 10 a 14, caracterizada porque el número de unidades de dosificación diaria que comprenden la combinación de Drospirenona y etinilestradiol es 21, 22, 23 ó 24 y porque el número de unidades de dosificación diaria que no contienen agente activo es 7, 6, 5 ó 4.	Rv. 1: (ver más arriba) Rv. 4: El método de acuerdo con la Rv. 3, donde la combinación progestina/estrógeno es administrada por 24 días por ciclo menstrual empezando sobre el primer día del ciclo menstrual. Rv.23: El método de acuerdo con la Rv.1, que comprende la administración de un placebo no-contraceptivo por 3-5 días consecutivos empezando sobre el día inmediatamente siguiente del último día de administración de la combinación progestina/estrógeno, tal que el total de días de administración de la combinación más el placebo no-contraceptivo es igual a 28 días. Rv. 29: Un kit contraceptivo adaptado para administración oral diaria, la cual comprende 28 unidades de dosificación separadas, 23-25 unidades de dosis separadas, dichas unidades de dosis cada una contiene una combinación de progestina en una dosis seleccionada del grupo que consiste de de 40-500µg trimegestone, 250µg- 4µg dienogest, y 250µg-4mg Drospirenona, y un estrógeno en una dosis diaria equivalente en actividad estrogénica a 10-20µg de etinilestradiol, donde cada unidad de dosificación contiene la misma dosificación de progestina y estrógeno; y 3-5 días de dichas unidades de dosificación contienen un placebo no-contraceptivo, en donde cada una de las 23-25 unidades de dosis contienen la misma dosificación de progestina y estrógeno.

De nuevo, una sencilla comparación entre los documentos citados lleva a concluir que la composición revelada en la solicitud en trámite, que comprende Drospirenona y Etinilestradiol carece por completo de novedad y de nivel inventivo puesto que la mencionada aplicación internacional WO 98/04269 ya revelaba la misma composición conformada por una progestina como Drospirenona y un estrógeno como Etinilestradiol en rangos como los que se pretende patentar en la solicitud que nos ocupa, pues en la primera el Etinilestradiol está presente en un rango comprendido entre 10-20µg (0.01 y 0.020mg), al paso que en la solicitud en trámite una dosis preferida del mismo principio activo es de 0.01mg hasta 0.05mg, preferiblemente 0.15 y 0.03mg, evidentemente comprendido en el rango anterior. Para el caso de Drospirenona, está presente en un rango comprendido entre 250µg y 4mg (0.25 y 4mg), valores que cobijan los rangos propuestos en la solicitud en trámite 2.0 a 4.0mg, preferiblemente 3.0 a 3.5mg.

Por lo anterior, no solo los rangos de dosificación propuestos para dichos compuestos "Etinilestradiol" y "Drospirenona" están revelados en la aplicación internacional WO 98/04269, al igual que el periodo de administración de los mismos, pues las formulaciones de **Etinilestradiol** y de **Drospirenona** se administran en unidades diarias de 28 ó de 24 y 4 unidades diarias con un compuesto no-contraceptivo, o 23 a 25 unidades de la combinación y 3 a 5 del compuesto no-contraceptivo.

1.7. La solicitud 00-065-489 carece de novedad y nivel inventivo por la materia revelada en el documento PCT WO 97/01342, publicado en 16/01/1997:

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 97/01342 presentada 27/06/1996 y publicada el 16/01/1997
Rv. 1: Una composición farmacéutica caracterizada por comprender, como primer agente activo, <u>Drospirenona</u> en una cantidad correspondiente a una dosis diaria, a ser administrada con la composición, desde aprox 2mg hasta 4mg, y, como segundo ingrediente activo, <u>etinilestradiol</u> en una cantidad correspondiente a una dosis diaria desde aprox 0.01mg hasta 0.05mg, junto con uno o más soportes o	Rv. 8: Preparación combinada de acuerdo con la Rv.1, donde el estrógeno del primer componente hormonal es seleccionado del grupo de: 17β estradiol, etinilestradiol y 17β-estradiol valerato y el gestágeno es seleccionado del grupo de compuestos: dienogest, gestodeno, levonorgestrel, desogestrel, 3-ketodesogestrel, <u>Drospirenona</u> (...) el estrógeno del segundo componente hormonal es seleccionado del

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 97/01342 presentada 27/06/1996 y publicada el 16/01/1997
excipientes farmacéuticamente aceptables, presentando dicha Drospirenona un área superficial mayor que 10.000cm²/g. Rv. 7: La composición de acuerdo con cualquiera de las reivindicaciones precedentes, caracterizada por comprender, una cantidad de <u>Drospirenona</u> correspondiente a una dosis desde aprox 3.0mg hasta 3.5mg, y <u>etinilestradiol</u> en una cantidad correspondiente a desde aprox 0.015mg hasta 0.03mg, en particular por comprender una cantidad de Drospirenona correspondiente a una dosis diaria de aproximadamente 3.0mg y etinilestradiol en una cantidad correspondiente a una dosis diaria de 0.03mg.	grupo de compuestos: 17β estradiol, <u>etinilestradiol</u> y 17β-estradiol valerato. Rv. 9: Preparación combinada de acuerdo con la Rv.1 donde el estrógeno del primer componente hormonal en cada unidad de dosis diaria contiene una dosis de: 1.0 a 6.0mg 17β estradiol, 0.015 a 0.025mg <u>etinilestradiol</u> y 1.0 a 4.0mg17β-estradiol valerato y el gestágeno en cada unidad de dosis diaria contiene una dosis de: 1.0 a 3.0mg dienogest, 0.05 a 0.075mg gestodeno, 0.05 a 0.125mg levonogestrel, 0.06 a 0.15mg desogestrel, 0.06 a 0.15mg 3-ketodesogestrel, 1.0 a 3.0mg <u>Drospirenona</u>.... Rv. 10: Preparación combinada de acuerdo con la Rv.1, en el que allí está contenido en cada dosis de unidad diaria en la segunda etapa, una cantidad de: 1.0 a 6.0mg 17β estradiol, 0.015 a 0.025mg <u>etinilestradiol</u> y 1.0 a 4.0mg17β-estradiol valerato.
Rv. 10: Una preparación farmacéutica, caracterizada por consistir de un número de unidades de dosificación.... y provistas para la administración oral durante <u>un período de 21 días consecutivos</u>..... Rv. 15: La preparación de acuerdo con cualquiera de las reivindicaciones 10 a 14, caracterizada por comprender adicionalmente 7 o menos unidades de dosificación diaria que no contienen agente activo, provistas para administración oral subsecuente al período de al menos 21 días consecutivos, siendo el número total de unidades de dosificación diaria <u>al menos 28</u> Rv. 16:	Rv. 1: La preparación farmacéutica combinada de dos etapas para la contracepción hormonal que contiene al menos 30 dosis de unidad diarias, en la cual, en su primera etapa, comprende como ingrediente hormonal activo una combinación de una preparación de estrógeno y, en una dosis que es suficiente para inhibir la ovulación, una preparación de gestágeno, en la forma de etapa sola y, en la segunda etapa comprende como ingrediente activo hormonal una preparación de estrógeno sólo, en el que la primera etapa comprende mínimo 25 y un máximo de 77 dosis de unidad diarias discretas o continuas y la segunda etapa comprende 5, 6 o 7 dosis de unidad diarias discretas o continuas, y en el que el número total de

Solicitud Colombiana 00-065-489 presentada 31/08/2000 y publicada 27/09/2002	PCT WO 97/01342 presentada 27/06/1996 y publicada el 16/01/1997
La preparación de acuerdo con cualquiera de las Rv.s 10 a 14, caracterizada porque el número de unidades de dosificación diaria que comprenden la combinación de Drospirenona y etinilestradiol es 21, 22, 23 ó 24m y porque el número de unidades de dosificación diaria que no contienen agente activo es 7, 6, 5 ó 4.	unidades diarias es igual al número total de los días del ciclo deseado de un mínimo de 30 y un máximo de 84 días. Rv. 2: Preparación combinada de acuerdo con la Rv.1, donde la primera etapa comprende 25 o 26 unidades de dosis diarias. Rv. 3: Preparación combinada de acuerdo con la Rv.1, donde la primera etapa comprende mínimo 28 y máximo 84 unidades de dosis diarias. Rv. 4: Preparación combinada de acuerdo con la Rv.3, donde la primera etapa comprende 28 unidades de dosis diarias. Rv. 5: Preparación combinada de acuerdo con la Rv.3, donde la primera etapa comprende 28 más 7, o 28 más un múltiplo de 7, unidades de dosis diarias. Rv. 6: Preparación combinada de acuerdo con la Rv.1, donde la segunda etapa comprende 7 unidades de dosis diarias. Rv. 7: Preparación combinada de acuerdo con la Rv.2, donde la segunda etapa comprende 5 o 6 unidades de dosis diarias, de modo que la preparación combinada tiene un total de 30 o 31 unidades de dosis diarias.

Como se observa, la comparación entre los documentos citados lleva a concluir que la composición revelada en la solicitud en trámite, que comprende Drospirenona y Etinilestradiol carece por completo de novedad y de nivel inventivo puesto que la mencionada aplicación internacional WO 97/01342 ya revelaba la misma composición conformada por un gestágeno como Drospirenona y un estrógeno como Etinilestradiol en rangos muy cercanos a los que se pretende patentar en la solicitud que nos ocupa, pues en la primera la Drospirenona está presente en un rango comprendido entre

1.0-3.0 mg, al paso que en la solicitud en trámite una dosis preferida del mismo principio activo es de 3.0mg, evidentemente comprendido en el rango anterior.

Para el caso de Etinilestradiol, está presente en un rango entre 0.015 y 0.025mg, valores que se encuentran dentro de los propuestos en la solicitud en trámite 0.01mg hasta 0.05mg.

Es importante resaltar que en la aplicación internacional WO 97/01342, se revelan periodo de administración de dichos principios activos, las formulaciones de **etinilestradiol** y de **Drospirenona**, se administran en unidades diarias de 28 ó de 25-26 y 5-6 unidades diarias del compuesto de la segunda etapa.

1.8. De igual manera, me permito presentar al Despacho apartes de un artículo de indudable relevancia para el tema que nos ocupa, pues el mismo demuestra una vez más la falta de Novedad y Nivel inventivo de la solicitud colombiana en trámite 00-065-489, veamos:

J Clin Endocrinol Metab. 1995 Jun;80(6):1816-21.

Effects of a new oral contraceptive containing an antimineralocorticoid progestogen, drospirenone, on the renin-aldosterone system, body weight, blood pressure, glucose tolerance, and lipid metabolism.
Oelkers W, Foidart JM, Dombrovicz N, Welter A, Heithecker R.

Division of Endocrinology, Klinikum Benjamin Franklin (Steglitz), Freie Universitat, Berlin, Germany.

Combined hormonal oral contraceptives (OCs) may lead to a mild rise in blood pressure and body weight. In rare instances, large increments in blood pressure are measured. We investigated the effect of a combination of ethinyl estradiol (EE) plus a progestogen with antimineralocorticoid, i.e. natriuretic, properties [Drospirenone (DRSP)] on body weight, blood pressure, the renin-aldosterone system, atrial natriuretic factor, plasma lipids, and glucose tolerance. It is anticipated that this will lead to the development of an OC that does not raise body weight or blood pressure. Four groups of 20 women each received 30 micrograms EE plus 3 mg DRSP (group A), 20 micrograms EE plus 3 mg DRSP (group B), 15 micrograms EE plus 3 mg DRSP (group C), and, as a control OC, 30 micrograms EE plus 150 micrograms levonorgestrel (Microgynon, Schering; group D) for 6 months. During the OC-free control cycles before and after treatment and throughout treatment, the target parameters were measured. Between the pretreatment cycle and the sixth treatment cycle, mean body weight fell by 0.8 to 1.7 kg in groups A, B, and C ($P < 0.05$ vs. D), whereas it rose by 0.7 kg in group D. Systolic and diastolic blood pressures fell by 1-4 mm Hg in groups A, B, and C (significant for A and C vs. D) and increased by 1-2 mm Hg in group D. Renin substrate rose equally in all groups ($P < 0.05$), whereas PRA and plasma aldosterone rose significantly only in the DRSP groups, presumably due to sodium loss. In the DRSP groups, high density lipoprotein cholesterol rose ($P < 0.05$), in contrast to group D. Low density lipoprotein cholesterol fell slightly ($P > 0.05$), whereas triglyceride levels showed a stronger increase in the DRSP groups ($P < 0.05$) than in group D. All groups attained good cycle control; group A had the best. Side-effects were minimal. To our

knowledge, this is the first report on a combined OC that leads to a small decrease in body weight and blood pressure. It may be especially beneficial for women susceptible for a gain in weight and a rise in blood pressure.

PIP: The potential of a new oral contraceptive (OC) containing drospirenone (DRSP) to avert the moderate increases in body weight and blood pressure often associated with use of existing combined OCs was investigated in a study of four groups of 20 German women each. Group A received 30 mcg of ethinyl estradiol (EE) and 3 mg of DRSP, Group B was administered 20 mcg of EE and 3 mg of DRSP, Group C received 15 mcg of EE and 3 mg of DRSP, and Group D was given a standard OC containing 30 mcg of EE and 150 mcg of levonorgestrel. Between the pretreatment cycle and the last (sixth) treatment cycle, mean body weight fell by 0.8-1.7 kg in Groups A, B, and C, but rose by 0.7 kg in Group D. Systolic and diastolic blood pressures fell by 1-4 mm Hg in Groups A, B, and C and rose by 1-2 mm Hg in Group D. Renin substrate rose equally in all four groups, while plasma renin activity, plasma aldosterone, and high density lipoprotein cholesterol rose significantly only in the three DRSP groups and serum triglyceride levels were significantly higher in Group D than in the three DRSP groups. Glucose tolerance increases were similar in all four groups. Finally, all groups--but especially Group A--experienced good cycle control and there were no serious side effects. These findings suggest that a combined OC containing DRSP may be especially beneficial for women who have a tendency to gain weight or experience a rise in blood pressure while taking OCs.

Publication Types:

- Clinical Trial
- Randomized Controlled Trial

PMID: 7775629 [PubMed - indexed for MEDLINE]

Luego de la lectura de este artículo publicado en **1995**, es necesario concluir que en esa época ya se sugería la combinación de anticonceptivos hormonales orales como es la combinación **de Etinilestradiol (EE) más un progestógeno como Drospirenona (DRSP)**. Además, es este artículo se revela las bondades de dicha combinación a saber: efectos secundarios mínimos, pequeña disminución de peso y de la tensión arterial siendo beneficioso para mujeres con una tendencia a ganar el peso o experimentar una subida de la tensión arterial.

De igual manera, se indican dosis tanto para Drospirenona como para Etinilestradiol, siendo -según el artículo- mejor la dosificación suministrada al grupo A que consiste de 30 µg de etinilestradiol (EE) y 3mg of DRSP; este rango de dosificación, así como los designados a los otros grupos de experimentación, se encuentran plenamente contemplados en el capítulo reivindicatorio de la solicitud colombiana en trámite 00-065-489, puesto que en él se revela que la dosis diaria a ser administrada de Drospirenona se encuentra desde 2mg hasta 4mg y para Etinilestradiol se encuentra desde 0.01mg hasta 0.05mg (10µg-50µg).

En consecuencia, las reivindicaciones 1, a 3, 6, 7, 10 a 12, 19 a 25, 29, 30, 34 a 36 y 39, incumplen los requisitos de patentabilidad en especial los contenidos en los artículos 16 y 18 de la Decisión 486.

Como petición especial en este punto, en caso de que el Despacho considere necesario contar con el texto completo del artículo en cuestión, ruego que en aplicación del artículo 46 de la Decisión 486 de 2000 se requiera al solicitante para que en el plazo legal allegue el documento mencionado a este expediente.

1.9. Ahora bien, si el fundamento de novedad de la solicitud colombiana en trámite estuviese dado por la presencia de Drospirenona en forma micronizada tal como lo revelan las reivindicaciones 2, 11, 24 y 26, me permito demostrar la carencia de nivel inventivo de la misma por obra de lo descrito en la publicación internacional WO 97/11680 publicada el **3 de abril de 1997**, en donde se revela de manera especial en las reivindicaciones 1, 11 y 17 un método de terapia de reemplazo hormonal (HRT) que comprende la administración de dosis sucesivas de un estrógeno y una pluralidad de dosis sucesivas de un progestágeno, donde el estrógeno es **etinilestradiol** o estradiol o un éster del mismo, estrona, estrona-sulfato o estrógenos conjugados y donde el progestágeno es progesterona micronizada, noretindrona o ésteres del mismo, norgestrel, clormadinona acetato, ciproterona acetato, desogestrel, 3-ketodesogestrel, **drospirenona**, norgestimate, levo-norgestrel o gestodeno.

Como se observa, esta publicación internacional no solo revela la combinación de Etinilestadiol y Drospirenona, sino que también sugiere la posibilidad de combinar el etinilestradiol con un compuesto progestágeno micronizado como la progesterona. Si bien es cierto el texto no se refiere de manera precisa a la presencia de Drospirenona en forma micronizada, si se puede generar la inquietud de experimentar con otros compuestos progestágenos en forma micronizada.

1.10. Por último me permito resaltar que tanto Drospirenona como Etinilestradiol son moléculas de las que se encuentran referencias por lo menos desde hace 40 años, veamos:

* La **DROSPIRENONA** está consignada en el Index Merck sexta edición - 1996 - en la página 585 bajo el numeral 3510, es interesante ver allí que aparece una patente en USA (4,129) desde **1978** para Schering AG. Este medicamento está consignado en las búsquedas bibliográficas inicialmente con el código ZK-30595 y

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su primera denominación genérica fue DIHIDROSPIRORENONE. En Med-Line se encuentran trabajos relacionados **a partir de 1982**.

* El **ETINILESTRADIOL** está consignado en el Index Merck sexta edición - 1996 - en la página 637 bajo el numeral 3780 y está referenciado en este documento que éste principio activo fue registrado como propiedad de Peht, Muller, Bull Soc. Chim. France, **desde 1951**.

De igual manera, es importante resaltar que este estrógeno ha sido tradicionalmente el que más se ha utilizado en diferentes combinaciones con diversos progestágenos, es así que desde los inicios de los anticonceptivos orales combinados se pueden encontrar combinaciones de etinilestradiol con otros principios activos con anterioridad a la fecha de prioridad de la presente solicitud:

- **Chlormadinone acetate:** Amenyl; Lutestral® (Cassenne); Menova (Merck KGaA)
- **Gestodene:** Femodene® (Schering AG); Femovan (Schering AG); Ginoden® (Schering AG); Harmonet® (Wyeth); Meliane® (Schering AG); Minulet® (Wyeth).
- **Lynestrenol:** Fysionorm (Nourypharma); Minilyn® (Organon); Ovanon® (Nourypharma); Ovoresta® (Organon); Yermonil (Novartis).
- **Medroxyprogesterone:** Provest® (Upjohn)
- **Megestrol acetate:** Co-Ervonum® (Glaxo); Kombiquens® (Novo); Noval® (Yamanouchi); Validan® (BDH; Searle).
- **Desogestrel:** Cyclosa® (Nourypharma); Dicromil® (Organon); Marvelon 150/30® (Akzo); Varnoline® (Organon); Desogen® (Organon): tablet oral-28 Desogestrel 0.15mg y Ethinyl Estradiol 0.03mg, Approval date Dec 10-1992 (Electronic Orange Book).
- **Ethinodiol:** Conova® (Glod Cross); Miniluteolas® (Serono); Demulen® (Searle): Tablet oral-21 Ethinyl Estradiol 0.035mg y Ethynidiol diacetate 1mg, Approval date prior to Jan 1-1982 (Electronic Orange Book)
- **Norethindrone:** Binovum® (Ortho-Cilag); Brevicon® (Syntex); Conceplan® (Grünenthal); Estrostep® (Warner-Lambert); Ovcon® (Bristol-Myers Squibb); Trinovum® (Ortho-Cilag); Nortrel® (Barr): Tablet oral-21 Ethinyl Estradiol 0.035mg y Norethindrone 0.5mg, Approval Feb 28-1992 (Electronic Orange Book).
- **Norgestrel:** Lo/Ovral® (Wyeth); Stédiril® (Wyeth); Ovral® (Wyeth): Tablet oral-28 Ethinyl Estradiol 0.05mg y Norgestrel 0.5mg, Approval prior to Jan 1-1982 (Electronic Orange Book).
- y ahora **Drospirenona:** Yasmin® (Schering AG)

Como vemos, queda demostrada una vez más que, el hecho de proponer una combinación de un estrógeno con un progestágeno- en el caso que nos ocupa Drospirenona-, carece por completo de nivel inventivo, pues, en el mercado se comercializan múltiples medicamentos que contienen Etinilestradiol junto a otros y distintos progestágenos como los enunciados anteriormente.

En conclusión y sin necesidad de ahondar en más pruebas, queda demostrado que carecen de novedad y nivel inventivo las reivindicaciones que conforman la solicitud 00-065-489, en la medida en que todas y cada una de sus características de la pretendida composición estaban comprendidas en el estado del arte antes de la fecha de presentación de la misma.

II. PRUEBAS

Para ilustración del Despacho, acompaño como anexos los siguientes:

1. Primera página y reivindicaciones de la aplicación internacional PCT WO 95/07081.
2. Primera página y reivindicaciones de la patente US 5,756,490
3. Primera página de la aplicación internacional PCT WO 95/26730.
4. Primera página y reivindicaciones de la aplicación internacional PCT WO 98/04267.
5. Primera página y reivindicaciones de la aplicación internacional PCT WO 98/27929.
6. Primera página y reivindicaciones de la patente US 5,583,129
7. Primera página de la aplicación internacional PCT WO 95/17194.
8. Primera página y reivindicaciones de la aplicación internacional WO 98/04269.
9. Primera página y reivindicaciones de la aplicación internacional WO 97/01342.
10. Primera página y reivindicaciones de la aplicación internacional WO 97/11680.

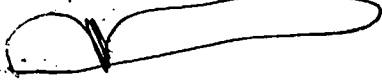
III. ANEXOS

1. Certificado de Existencia y Representación de la sociedad **LAFRANCOL S.A.**, expedido por la Cámara de Comercio de Cali.
2. Poder conferido al suscrito por **LAFRANCOL S.A.**, para la atención de asuntos relacionados con Propiedad Industrial.

IV. NOTIFICACIONES

LAFRANCOL S.A. las recibirá en la Secretaría de ese Despacho o en su dirección de notificaciones judiciales, ubicada en la Carrera 1a. No. 46-84 de la ciudad de Cali, departamento del Valle del Cauca. El suscrito apoderado las recibirá en la misma Secretaría, o en mi oficina de abogado, ubicada en la Carrera 17 No. 88-23, Oficina 205, Fax (1) 621.25.42 de Bogotá, D.C.

Señor Superintendente,



José Luis Reyes Villamizar
C.C. 79'152.473 de Usaquén
T.P.A. 44.655 de C.S.J.



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REPUBLICA DE COLOMBIA

NOTARIA 33 DE BOGOTA D.C.

ESCRITURA PUBLICA NUMERO: 1 6 5 8

- - MIL SEISCIENTOS CINCUENTA Y OCHO - -

FECHA DE OTORGAMIENTO: CUATRO (4) DE

JULIO - - - - -

DEL AÑO DOS MIL DOS (2002). -----

ACTO: -----

1.- PODER ESPECIAL.-----

OTORGADO POR: LAFRANCOL S.A., con Nit. No. - - -

890.301.463-8 - - - - -

a favor de: JOSE LUIS REYES VILLAMIZAR, con C.C.No.

79.152.473 de Usaquén - Bogotá.-----

DIANA BEATRIZ LOPEZ

NOTARIA TREINTA Y TRES (33) DE BOGOTA D.C.

EN LA CIUDAD DE BOGOTA, DISTRITO CAPITAL,

DEPARTAMENTO DE CUNDINAMARCA, REPUBLICA DE COLOMBIA,

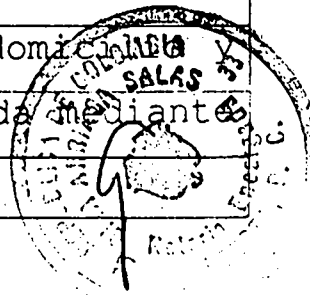
ANTE MI, DIANA BEATRIZ LOPEZ, - - - - -

NOTARIA TREINTA Y TRES (33) - - - - -

DE ESTE CIRCULO SE OTORGA LA ESCRITURA PUBLICA QUE
SE CONSIGNA EN LOS SIGUIENTES TERMINOS:-----COMPARECIO: JUAN MARIA RENDON, mayor de edad,
vecino de Bogotá, D.C, identificado con la cédula de
ciudadanía número 17.125.100 - - - - -

Expedida en Bogotá - - - - -

Y manifestó:-----

PRIMERO: Que obra en su carácter de Representante
legal de en nombre y representación de LAFRANCOL
sociedad comercial organizada y existente
a las leyes colombianas, con domicilio
sede principal en Cali - Valle, constituida mediante
escritura pública número

De la Notaría
De fecha
Reformada en distintas oportunidades, según consta en el certificado expedido por la Cámara de Comercio de Cali - Valle.-----
SEGUNDO: Que en tal carácter, por este público instrumento, concede PODER ESPECIAL, PERO AMPLIO Y SUFICIENTE a JOSE LUIS REYES VILLAMIZAR , mayor de edad, vecino de Bogotá, D.C, identificado con la cédula de ciudadanía número 79.152.473 de Usaquén, abogado en ejercicio, portador de la T.P.No. 446 55 del C.S. de la J., domiciliado en Bogotá, D.C, República de Colombia, con dirección en la carrera 17 No. 88-23, Oficina 205, de la misma ciudad, como su representante con poder especial, amplio y suficiente, para obtener la protección los derechos de propiedad industrial y de libre competencia de Lafrancol S.A., a cuyo efecto lo autorizo, par que nos represente ante cualesquiera entidad o persona, ejerza o no jurisdicción, especialmente ante las el orden administrativo, contencioso, administrativo y judicial, en todo lo concerniente a los siguientes asuntos:-----
a.- Obtención, formulación de oberservaciones y oposición a patentes de invención, modelos y dibujos industriales y el registro de licencias de uso de esos derechos.-----
b.- Las acciones de oposición, cancelación, nulidad, medidas cautelares, concesión de licencias y en general los procesos civiles, penales, administrativos o contencioso- administrativos relacionados con estos derechos u otros de diferente naturaleza, acciones y procesos que



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promueva Lafrancol S.A., o se promuevan contra esta y.-----

c.- Para que en todos los asuntos arriba determinados, tanto en los términos señalados por los artículos 42 y complementarios

de la decisión 486 de la Comunidad Andina como del artículo 70 del C.P.C. pueda sustituir este mandato, transigir, conciliar, admitir los hechos del proceso, desistir, cancelar, recibir, renunciar y ratificar los actos de agentes oficiosos y ceder a título gratuito y/o oneroso, licenciar, inscribir contratos de cesión y de licencia, inscripción de prendas, levantamiento de prendas, inscribir derechos de retención y en general todos los asuntos que se deban tramitar ante la Superintendencia de Industria y Comercio.-----

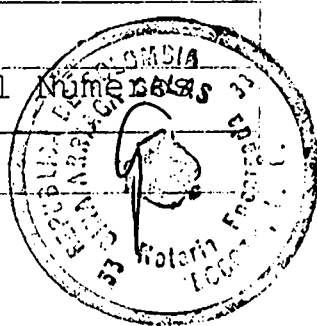
Manifestamos que de la veracidad de las declaraciones del contenido y autenticidad de los documentos anexos en éste instrumento público sólo respondemos los comparecientes.-

HASTA AQUI LA MINUTA PRESENTADA

OTORGAMIENTO Y AUTORIZACION: Leído el presente instrumento Público por los comparecientes y advertidos de la formalidad de su contenido, dentro del término legal, lo hallaron conforme con sus intenciones y lo aprobaron en todas sus partes y firmaron junto con la Suscrita Notaria quien da fe y autoriza. -----

Se utilizaron las hojas de papel Notarial AA 8567146, 8567152.

22 JUN 2004
GUSTAVO SAMPER RODRIGUEZ
NOTARIO LEYNTSES



DERECHOS NOTARIALES: \$ 49.600.00

RESOLUCION No. 4188 del 28 de Diciembre del 2001

Juan Maria Rendon
JUAN MARIA RENDON

C.C. No. 17.125.100 de Bogotá

LAFRANCOL S.A.

Nit. No. 890.309.463-8

TEL: 622-04-66.

ACEPTO :

Jose Luis Reyes Villamizar
JOSE LUIS REYES VILLAMIZAR

C.C. No. 79.152.473

TEL: 621-2631

Diana Beatriz Lopez
DIANA BEATRIZ LOPEZ

DIANA BEATRIZ LOPEZ

NOTARIA 33 DE BOGOTA D.C.

n.c.a.

Es segunda COPIA (FOTOCOPIA)

tomada de su original que se expide conforme al

Artículo 79 del Decreto 960 de 1970 en 06

hojas útiles con destino a Jose Luis Reyes

Villamizar

Santafé de Bogotá D.C.

SEDA: 1772/79

05 JUL. 2002

Qui Cumme



FECHA: JUEVES 03-MARZO-2005 11:41:49

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REPUBLICA DE COLOMBIA
CERTIFICADO DE EXISTENCIA Y REPRESENTACION

EL SUSCRITO SECRETARIO DE LA CAMARA DE COMERCIO DE CALI

CERTIFICA

NOMBRE: LABORATORIO FRANCO COLOMBIANO LAFRANCOL S.A.
DOMICILIO: CALI VALLE
DIRECCION COMERCIAL: K-1 46-84
DIRECCION NOTIFICACION JUDICIAL: K-1 46-84
CIUDAD: CALI
MATRICULA MERCANTIL NRO. 5233-14 FECHA MATRICULA: 02 DE OCTUBRE DE 1958
DIRECCION ELECTRONICA: ech-compras@lafrancol.com y miguelozada@lafrancol.com
AFILIADO

NIT: 890301463-8

CERTIFICA

CERTIFICA

CERTIFICA

QUE POR ESCRITURA NRO. 1434 DEL 29 DE SEPTIEMBRE DE 1958 NOTARIA CUARTA DE CALI
INSCRITA EN LA CAMARA DE COMERCIO EL 02 DE OCTUBRE DE 1958 BAJO EL NRO. 18319 DEL
LIBRO IX SE CONSTITUYO LA SOCIEDAD DENOMINADA LABORATORIO FRANCO COLOMBIANO LAFRANCOL
S.A.

REFORMAS

DOCUMENTO	FECHA DOC	ORIGEN	FECHA INS	NRO INS	LIBRO
E.P. 1104	30/06/1959	NOTARIA CUARTA DE CALI	07/07/1959	19467	
E.P. 2309	26/12/1959	NOTARIA CUARTA DE CALI	29/12/1959	20089	
E.P. 1725	24/07/1962	NOTARIA CUARTA DE CALI	26/07/1962	24528	
E.P. 1824	31/07/1963	NOTARIA CUARTA DE CALI	14/08/1963	26360	
E.P. 2422	25/10/1963	NOTARIA CUARTA DE CALI	28/10/1963	26717	
E.P. 1720	16/09/1964	NOTARIA CUARTA DE CALI	22/09/1964	28406	
E.P. 101	31/03/1966	NOTARIA CUARTA DE CALI	01/02/1966	31039	
E.P. 552	21/03/1968	NOTARIA CUARTA DE CALI	01/04/1968	35711	
E.P. 4964	12/12/1972	NOTARIA CUARTA DE CALI	16/01/1973	2975	IX
E.P. 1956	30/05/1973	NOTARIA CUARTA DE CALI	13/06/1973	4337	IX
E.P. 4964	12/12/1972	NOTARIA CUARTA DE CALI	28/11/1973	5994	IX
E.P. 5579	12/11/1973	NOTARIA CUARTA DE CALI	28/11/1973	5995	IX
E.P. 4406	30/09/1974	NOTARIA CUARTA DE CALI	28/11/1974	10887	IX
E.P. 192	03/02/1978	NOTARIA CUARTA DE CALI	14/02/1978	25355	IX
E.P. 3479	07/09/1979	NOTARIA CUARTA DE CALI	19/09/1979	34204	IX
E.P. 4274	12/12/1984	NOTARIA NOVENA DE CALI	19/12/1984	73088	IX
E.P. 1155	21/05/1985	NOTARIA SEPTIMA DE CALI	03/06/1985	76917	IX
E.P. 1553	20/06/1986	NOTARIA SEPTIMA DE CALI	08/07/1986	85997	IX
E.P. 1458	09/06/1987	NOTARIA SEPTIMA DE CALI	24/12/1987	3508	IX
E.P. 7222	14/12/1988	NOTARIA TERCERA DE CALI	19/12/1988	13847	IX
E.P. 4530	04/09/1989	NOTARIA TERCERA DE CALI	07/09/1989	21508	IX
E.P. 3865	02/08/1990	NOTARIA TERCERA DE CALI	13/08/1990	31708	IX
E.P. 8305	13/12/1995	NOTARIA TERCERA DE CALI	23/01/1996	557	IX
E.P. 4559	16/11/1999	NOTARIA TERCERA DE CALI	26/11/1999	7911	IX
E.P. 67004	02/12/2004	NOTARIA TERCERA DE CALI	14/12/2004	13371	IX

CERTIFICA

VIGENCIA: 29 DE SEPTIEMBRE DEL AÑO 2057

CERTIFICA

OBJETO SOCIAL: SE OCUPA EN: A) FABRICACION, DISTRIBUCION Y VENTA EN EL TERRITORIO DE LA REPUBLICA DE COLOMBIA O EN EL EXTRANJERO DE CUALESQUIERA TIPOS DE VACUNAS, PRODUCTOS QUIMICOS, FARMACEUTICOS, ALIMENTICIOS, FUNCIONALES, COSMETICOS Y FISIOLOGICOS, REACTIVOS, ANTIBIOTICOS PARA USO HUMANO Y VETERINARIO, PRODUCTOS DIFETICOS, COLORANTES DE PERFUMERIA Y AFINES. B) LA REPRESENTACION DE CASAS FABRICANTES NACIONALES O EXTRANJERAS DE ESTOS MISMOS PRODUCTOS. C) FABRICACION, IMPORTACION, DISTRIBUCION Y VENTA DE CUALESQUIERA TIPOS DE PRODUCTOS VETERINARIOS. D) LA REALIZACION DE TODOS LOS ACTOS, CONTRATOS, CONVENIOS, DIRECTAMENTE RELACIONADOS CON LOS FINES ANTES INDICADOS EN DESARROLLO DE SU OBJETO SOCIAL PODRA LA SOCIEDAD ADQUIRIR Y VENDER BIENES RAICES Y MUEBLES, DARLOS Y TOMARLOS EN ARRENDAMIENTO, GARANTIZAR CON HIPOTECA SOBRE LOS BIENES RAICES O CON PRENDA SOBRE LOS BIENES MUEBLES, OBLIGACIONES A CARGO DE LA SOCIEDAD, DAR Y RECIBIR A MUTUO COMERCIAL DINERO Y CELEBRAR LOS CONTRATOS COMERCIALES DE CAMBIO EN TODAS SUS MANIFESTACIONES, FUNDAR EMPRESAS QUE TENGAN LA MISMA FINALIDAD SOCIAL O ASOCIARSE CON ELLAS.

CERTIFICA

ORGANOS DE DIRECCION Y ADMINISTRACION: 1) ASAMBLEA GENERAL DE ACCIONISTAS; 2) JUNTA DIRECTIVA; 3) GERENCIA GENERAL. TAMBIEN TENDRA UN REVISOR FISCAL.

ADMINISTRACION: LA ADMINISTRACION INMEDIATA DE LA SOCIEDAD, SU REPRESENTACION LEGAL Y LA GESTION DE LOS NEGOCIOS SOCIALES ESTARAN A CARGO DE UN GERENTE, DESIGNADO POR LA ASAMBLEA GENERAL DE ACCIONISTAS, PARA PERIODOS DE DE DOS (2) AÑOS, REELEGIBLE INDEFINIDAMENTE Y REMOVIBLE LIBREMENTE POR ELLA EN CUALQUIER TIEMPO.

PARAGRAFO PRIMERO: SUPLENTE: EL GERENTE Y REPRESENTANTE LEGAL TENDRA LOS SIGUIENTES SUPLENTE: PRIMERO, SEGUNDO Y TERCERO QUE LO REEMPLAZARAN EN LOS CASOS DE AUSENCIAS ACCIDENTAL, TEMPORAL Y ABSOLUTA CON LAS LIMITACIONES QUE MAS ADELANTE SE ESTABLECEN.

PARAGRAFO SEGUNDO: EL TERCER SUPLENTE DEL GERENTE GENERAL Y REPRESENTANTE LEGAL ESTARA LIMITADO EN SUS FUNCIONES A: COMPROMETER LOS NEGOCIOS SOCIALES EN CUANTIA NO SUPERIOR A CUATRO MIL DOSCIENTOS SALARIOS MINIMOS MENSUALES LEGALES VIGENTES (4.200 SMMVLV); CELEBRAR CONTRATOS COMERCIALES DE COMPRAVENTA EN GENERAL YA SEA CON LA NACION (MINISTERIOS Y DEPARTAMENTOS ADMINISTRATIVOS), ENTIDADES DESCENTRALIZADAS (ESTABLECIMIENTOS PUBLICOS), EMPRESAS INDUSTRIALES Y COMERCIALES DEL ESTADO, SOCIEDADES DE ECONOMIA MIXTA), EMPRESAS SOCIALES CIVILES Y COMERCIALES, CUALQUIERA QUE SEA SU NATURALEZA CON FACULTADES PARA EXIGIR O SUSCRIBIR LOS DOCUMENTOS Y GARANTIAS NECESARIOS HASTA LA CUANTIA ANTES MENCIONADA.

FUNCIONES DEL GERENTE: A) B) EJECUTAR Y HACER CUMPLIR LOS ACUERDOS Y DECISIONES DE LA ASAMBLEA GENERAL DE ACCIONISTAS Y DE LA JUNTA DIRECTIVA Y HACER USO DE LA RAZON SOCIAL. C) REPRESENTAR LEGALMENTE A LA SOCIEDAD CONFORME A LA LEY ANTE TODAS LAS AUTORIDADES CIVILES, JUDICIALES, MILITARES, SANITARIAS, DE POLICIA, ADUANALES, POSTALES, ETC. G) LLEVAR A CABO LA GESTION COMERCIAL Y FINANCIERA. H) TENDRA LA PLENA RESPONSABILIDAD DE LA ACCION ADMINISTRATIVA, LA COORDINACION Y LA SUPERVISION GENERAL DE LA SOCIEDAD, LAS CUALES CUMPLIRA CON ARREGLO A LAS NORMAS DE LOS ESTATUTOS. EN EL EJERCICIO DE SUS FUNCIONES, EL GERENTE PUEDE: 1. COMPARECER EN LOS JUICIOS, TRANSIGIR Y COMPROMETER LOS NEGOCIOS SOCIALES DE CUALQUIERA NATURALEZA QUE FUEREN, INTERPONER TODO GENERO DE RECURSOS Y CONSTITUIR APODERADOS QUE LO REPRESENTEN JUDICIAL O EXTRAJUDICIALMENTE. 2. ENAJENAR A CUALQUIER TITULO Y COMPRAR BIENES MUEBLES, CELEBRAR CONTRATOS COMERCIALES DE COMPRA Y VENTA EN GENERAL YA SEA CON LA NACION (MINISTERIOS Y DEPARTAMENTOS ADMINISTRATIVOS), ENTIDADES DESCENTRALIZADAS (ESTABLECIMIENTOS PUBLICOS), EMPRESAS INDUSTRIALES Y COMERCIALES DEL ESTADO, SOCIEDADES DE ECONOMIA MIXTA), EMPRESAS O SOCIEDADES CIVILES Y COMERCIALES CUALQUIERA QUE SEA SU NATURALEZA Y SU CUANTIA, CON AMPLIAS FACULTADES QUE EXIGIR O SUSCRIBIR LOS DOCUMENTOS, Y GARANTIAS NECESARIAS. CONTRATAR PRESTAMOS CUALQUIERA QUE SEA SU CUANTIA, CELEBRAR EL CONTRATO COMERCIAL DE MUTUO EN TODAS SUS MANIFESTACIONES Y SUSCRIBIR LOS DOCUMENTOS Y

C.C.C. 11090804

COMPROMISOS RESPECTIVOS: HACER DEPOSITOS EN BANCOS Y AGENCIAS BANCARIAS, FIRMAR CHEQUES, LETRAS, PAGARES, GIROS, LIBRANZAS Y CUALQUIER OTRO DOCUMENTO, ASI COMO NEGOCIAR ESTOS INSTRUMENTOS, TENERLOS, COBRARLOS, PAGARLOS, DESCARGARLOS, ETC.

CERTIFICA

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DOCUMENTO: ACTA No. 99 DEL 14 DE MARZO DE 2002

ORIGEN: ASAMBLEA GENERAL DE ACCIONISTAS

INSCRIPCION: 09 DE SEPTIEMBRE DE 2002 No. 14675 DEL LIBRO IX

FUE(ON) NOMBRADO(S):

VICEPRESIDENTE

ROBERTO S. VENTURA PAULY

C.C.C. 16697614

PRESIDENTE

ESTHER ISABEL VENTURA DE RENDON

C.C.C. 38979831

GERENTE GENERAL Y REPRESENTANTE LEGAL

JUAN MARIA RENDON GUTIERREZ

C.C.C. 17425100

PRIMER SUPLENTE DEL GERENTE

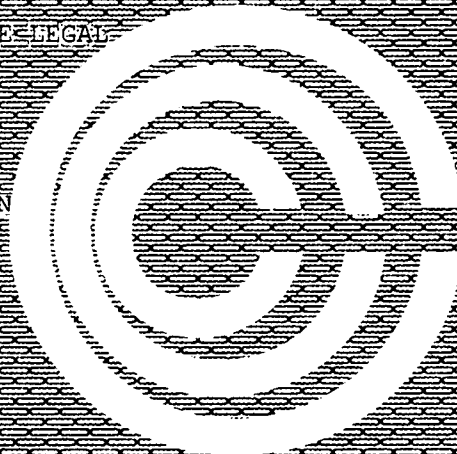
ESTHER ISABEL VENTURA DE RENDON

C.C.C. 38979831

SEGUNDO SUPLENTE DEL GERENTE

ROBERTO M. VENTURA CRISPINO

C.C.C. 9086003



CERTIFICA

DOCUMENTO: ACTA No. 105 DEL 09 DE NOVIEMBRE DE 2004

ORIGEN: ASAMBLEA DE ACCIONISTAS

INSCRIPCION: 17 DE FEBRERO DE 2005 No. 2032 DEL LIBRO IX

FUE(ON) NOMBRADO(S):

TERCER SUPLENTE DEL GERENTE

JUAN PABLO REYES VILLAMIZAR

C.C.C. 80419709

CERTIFICA

DOCUMENTO: ACTA No. 99 DEL 14 DE MARZO DE 2002

ORIGEN: ASAMBLEA GENERAL DE ACCIONISTAS

INSCRIPCION: 09 DE SEPTIEMBRE DE 2002 No. 14675 DEL LIBRO IX

DOCUMENTO: ACTA No. 105 DEL 09 DE NOVIEMBRE DE 2004

ORIGEN: ASAMBLEA DE ACCIONISTAS

INSCRIPCION: 17 DE FEBRERO DE 2005 No. 2031 DEL LIBRO IX

FUE(ON) NOMBRADO(S):

JUNTA DIRECTIVA

PRINCIPALES

PRIMER RENGLON

ESTHER ISABEL VENTURA DE RENDON
C.C. 38979831

SEGUNDO RENGLON

ROSA VICTORIA VENTURA DE VISBAL
C.C. 38992048

TERCER RENGLON

JUAN PABLO REYES VILLAMIZAR
C.C. 80419709

SUPLENTE

PRIMER RENGLON

JUAN MARIA RENDON GUTIERREZ
C.C. 17125100

SEGUNDO RENGLON

ROBERTO M. VENTURA CRISPINO
C.C. 9086003

TERCER RENGLON

ROBERTO S. VENTURA PAULY
C.C. 16697614



DOCUMENTO: ACTA No. 99 DEL 14 DE MARZO DE 2002
ORIGEN: ASAMBLEA GENERAL DE ACCIONISTAS
INSCRIPCION: 09 DE SEPTIEMBRE DE 2002 No. 14675 DEL LIBRO IX

FUE(RON) NOMBRADO(S):

REVISOR FISCAL

ALFREDO LOPEZ & CIA. LIMITADA
NIT 800026893

**CAMARA
DE COMERCIO
DE CALI**

CERTIFICA

DOCUMENTO: DOCUMENTO PRIVADO DEL 30 DE AGOSTO DE 2002
ORIGEN: ALFREDO LOPEZ & CIA. LTDA
INSCRIPCION: 09 DE SEPTIEMBRE DE 2002 No. 14678 DEL LIBRO IX

FUE(RON) NOMBRADO(S):

REVISOR FISCAL PRINCIPAL

ALFREDO LOPEZ CAMPO
C.C. 8241912

REVISOR FISCAL SUPLENTE

YAMILEG OSPINA MORALES
C.C. 31996911

CCC11090805

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CERTIFICA

CAPITAL AUTORIZADO: \$1,500,000,000
 NUMERO DE ACCIONES: 150,000,000
 VALOR NOMINAL: \$10
 CAPITAL SUSCRITO: \$903,756,830
 NUMERO DE ACCIONES: 90,375,683
 VALOR NOMINAL: \$10
 CAPITAL PAGADO: \$903,756,830
 NUMERO DE ACCIONES: 90,375,683
 VALOR NOMINAL: \$10

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CERTIFICA

QUE EL 03 DE MAYO DE 2004 BAJO LOS NROS. 4859, 4860 Y 4861 DEL LIBRO IX, SE INSCRIBIO EN LA CAMARA DE COMERCIO UN DOCUMENTO PRIVADO DE FECHA ABRIL 30 DE 2004 RELATIVO A LA DECLARACION DE UN GRUPO EMPRESARIAL, EN EL CUAL CONSTA:

CONTROLANTE: LABORATORIO FRANCO COLOMBIANO LAFRANCOL S.A.
 DOMICILIO: CALI
 NACIONALIDAD: COLOMBIANA

ACTIVIDAD: A) LA FABRICACION, DISTRIBUCION Y VENTA EN EL TERRITORIO DE LA REPUBLICA DE COLOMBIA O EN EL EXTRANJERO DE CUALESQUIERA TIPOS DE VACUNAS, PRODUCTOS QUIMICOS, FARMACEUTICOS Y FISIOLÓGICOS, REACTIVOS, ANTIBIOTICOS PARA USO HUMANO Y VETERINARIO, PRODUCTOS DIETETICOS, COLORANTES DE PERFUMERIA Y AFINES. B) LA REPRESENTACION DE CASAS FABRICANTES NACIONALES O EXTRANJERAS DE ESTOS MISMOS PRODUCTOS. C) FABRICACION, IMPORTACION, DISTRIBUCION Y VENTA DE CUALESQUIERA TIPOS DE PRODUCTOS VETERINARIOS. D) LA REALIZACION DE TODOS LOS ACTOS, CONTRATOS, CONVENIOS, DIRECTAMENTE RELACIONADOS CON LOS FINES ANTES INDICADOS. EN DESARROLLO DE SU OBJETO SOCIAL PUEDE LA SOCIEDAD ADQUIRIR Y VENDER BIENES RAICES Y MUEBLES, DARLOS Y TOMARLOS EN ARRENDAMIENTO, GARANTIZAR CON HIPOTECA SOBRE LOS BIENES RAICES O CON PRENDA SOBRE LOS BIENES MUEBLES, OBLIGACIONES A CARGO DE LA SOCIEDAD, DAR Y RECIBIR A MUTUO COMERCIAL DINERO Y CELEBRAR LOS CONTRATOS COMERCIALES DE CAMBIO EN TODAS SUS MANIFESTACIONES, FUNDAR EMPRESAS QUE TENGAN LA MISMA FINALIDAD SOCIAL O ASOCIARSE CON ELLAS.

DE ACUERDO CON EL NUMERAL 3, ARTICULO 27 DE LA LEY 222 QUE SE TRANSCRIBE: CUANDO LA MATRIZ, DIRECTAMENTE O POR INTERMEDIO O CON EL CONCURSO DE LAS SUBORDINADAS, EN RAZON DE UN ACTO O NEGOCIO CON LA SOCIEDAD CONTROLADA O CON SUS SOCIOS, EJERZA INFLUENCIA DOMINANTE EN LAS DECISIONES DE LOS ORGANOS DE ADMINISTRACION DE LA SOCIEDAD.

DE COMERCIO
 DE CALI

SUBORDINADA: LABORATORIOS PAULY PHARMACEUTICAL S.A.
 NIT: 8007097402-6
 DOMICILIO: CALI

ACTIVIDAD PRINCIPAL: DISTRIBUCION Y COMERCIALIZACION DE PRODUCTOS FARMACEUTICOS DE USO HUMANO.

PRESUPUESTO DE CONTROL: NUMERAL 3, ARTICULO 27 DE LA LEY 222.

SUBORDINADA: AMERICAN GENERICS S.A.
 NIT: 8007223214-9
 DOMICILIO: CALI

ACTIVIDAD PRINCIPAL: DISTRIBUCION Y COMERCIALIZACION DE PRODUCTOS FARMACEUTICOS DE USO HUMANO.

PRESUPUESTO DE CONTROL: NUMERAL 3, ARTICULO 27 DE LA LEY 222.

SUBORDINADA: LAFRANCOL INTERNACIONAL S.A.
 NIT: 8157003912-2

DOMICILIO: ZONA FRANCA DEL PACIFICO BODEGA 6 PISO 2 KM 6 VIA YUMBO AEROPUERTO PALMIRA
 ACTIVIDAD PRINCIPAL: REALIZACION DE ACTIVIDADES DESDE LA ZONA FRANCA, LAS CUALES CONSISTEN EN LA PRESTACION DE TODO TIPO DE SERVICIOS, ASI COMO DISTRIBUIR, VENDER, COMPRAR, IMPORTAR, EXPORTAR Y ALMACENAR TODO TIPO DE MATERIAS PRIMAS, PRODUCTOS

TERMINADOS Y SEMITERMINADOS RELACIONADOS CON LA INDUSTRIA DE ALIMENTOS, COSMETICA Y FARMACEUTICA.

PRESUPUESTO DE CONTROL: LABORATORIO FRANCO COLOMBIANO LATRANCOL S.A. POSEE EL 403 Y LABORATORIOS PAULY PHARMACEUTICAL S.A. POSEE EL 403 Y AMERICAN GENERICS S.A. POSEE EL 19-973.

SUBORDINADA: LATRANCOL PERU S.A.

REGISTRO UNICO DE CONTRIBUYENTE: 20504784089

DOMICILIO: AVENIDA JAVIER PRADO OESTE 829 MACDAGENA, LIMA 17

ACTIVIDAD PRINCIPAL: DISTRIBUCION Y COMERCIALIZACION DE PRODUCTOS FARMACEUTICOS DE USO HUMANO.

PRESUPUESTO DE CONTROL: LABORATORIO FRANCO COLOMBIANO LATRANCOL S.A. POSEE EL 93-08.

SUBORDINADA: LATRANCOL (USA) CORP.

NUMERO DE DOCUMENTO: P-00000090126

DOMICILIO: 2121 PONCE DE LEON BLVD. STE 240 CORAL GABLES FL 33134

ACTIVIDAD PRINCIPAL: DISTRIBUCION Y COMERCIALIZACION DE PRODUCTOS FARMACEUTICOS DE USO HUMANO.

PRESUPUESTO DE CONTROL: LABORATORIO FRANCO COLOMBIANO LATRANCOL POSEE EL 65-258.

CERTIFICA

QUE A NOMBRE DE LA SOCIEDAD FIGURA MATRICULADO EN LA CAMARA DE COMERCIO BAJO EL NRO. 5234-2 ESTABLECIMIENTO DE COMERCIO: LABORATORIO FRANCO COLOMBIANO LATRANCOL UBICADO EN: K 1 46 84 DE CALI RENOVO: POR EL AÑO 2004

CERTIFICA

QUE A NOMBRE DE LA SOCIEDAD FIGURA MATRICULADO EN LA CAMARA DE COMERCIO BAJO EL NRO. 369809-2 ESTABLECIMIENTO DE COMERCIO: LABORATORIO FRANCO COLOMBIANO LATRANCOL S.A UBICADO EN: CRA. 1 NRO. 46A 70 DE CALI FECHA MATRICULA: 29 DE ABRIL DE 1994 RENOVO: POR EL AÑO 2004



QUE A NOMBRE DE LA SOCIEDAD FIGURA MATRICULADO EN LA CAMARA DE COMERCIO BAJO EL NRO. 369810-2 ESTABLECIMIENTO DE COMERCIO: LABORATORIO FRANCO COLOMBIANO LATRANCOL S.A UBICADO EN: K 1 46 32 DE CALI FECHA MATRICULA: 29 DE ABRIL DE 1994 RENOVO: POR EL AÑO 2004

CERTIFICA

QUE LA SOCIEDAD EFECTUO LA RENOVACION DE SU MATRICULA MERCANTIL EL 25 DE MARZO DE 2004

CERTIFICA

QUE NO FIGURAN OTRAS INSCRIPCIONES QUE MODIFIQUEN TOTAL O PARCIALMENTE EL PRESENTE CERTIFICADO.

LOS ACTOS Y DOCUMENTOS REGISTRADOS QUEDAN EN FIRME CINCO (5) DIAS HABILDES DESPUES DE LA FECHA DE SU PUBLICACION EN EL BOLETIN NOTICIAS DE INSCRIPCIONES, SIEMPRE Y CUANDO DENTRO DE DICHO TERMINO NO SEAN OBJETO DE RECURSOS EN LA VIA GUBERNATIVA.

DE CONFORMIDAD CON EL DECRETO 2150 DE 1995 Y LA AUTORIZACION IMPARTIDA POR LA SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO DA FIRMA MECANICA QUE APARECE A CONTINUACION TIENE PLENA VALIDEZ PARA TODOS LOS EFECTOS LEGALES

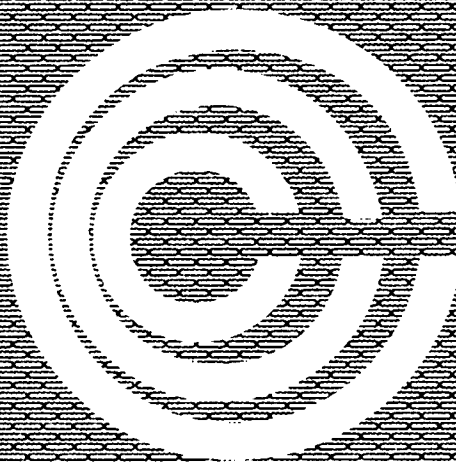
CCC11090806

DADO EN CALI A LOS 03 DIAS DEL MES DE MARZO

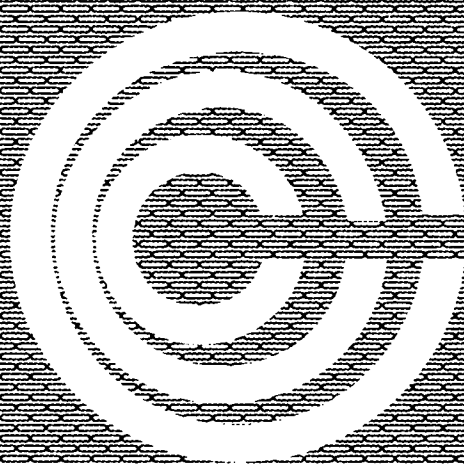
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EL SECRETARIO *[Handwritten Signature]*



**CAMARA
DE COMERCIO
DE CALI**



**CAMARA
DE COMERCIO
DE CALI**

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PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶: A61K 31/57, 31/565		A1	(11) International Publication Number: WO 95/07081 (43) International Publication Date: 16 March 1995 (16.03.95)
(21) International Application Number: PCT/EP94/02997 (22) International Filing Date: 8 September 1994 (08.09.94) (30) Priority Data: 9301562 9 September 1993 (09.09.93) NL (71) Applicant (for all designated States except US): SATURNUS AG [LU/LU]; 11, rue Aldringen, L-2960 Luxembourg (LU). (72) Inventor; and (75) Inventor/Applicant (for US only): KONINCKX, Philippe, Robert, Marie, Wilhelmus, Ghislain [BE/BE]; Vuilenbos 2, B-3360 Bierbeek (BE). (74) Agent: PRINS, Hendrik, Willem; Arnold & Siedsma, Sweelinckplein 1, NL-2517 GK The Hague (NL).		(81) Designated States: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, ES, FI, GB, GE, HU, JP, KE, KG, KP, KR, KZ, LK, LT, LU, LV, MD, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SI, SK, TJ, TT, UA, US, UZ, VN, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG), ARIPO patent (KE, MW, SD). Published <i>With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>	
(54) Title: PREPARATION FOR SUBSTITUTION THERAPY, CONTAINING AT LEAST ONE PROGESTOGEN AND AT LEAST ONE ESTROGEN (57) Abstract The invention relates to a preparation for substitution therapy and oral contraception comprising at least one progestogen and at least one estrogen in which the estrogen dose varies with a periodicity such that blood loss is substantially avoided, wherein the periodicity is preferably less than 10 days, more preferably less than 7 days, such as preparations containing the progestogen and/or estrogen in an oral, transdermal, parenteral and/or implantable application form.			

CLAIMS

1. Preparation for substitution therapy and for oral contraception comprising at least one progestogen and at least one estrogen, in which the estrogen dose varies with a periodicity such that blood loss is substantially avoided.

5 2. Preparation as claimed in claim 1, wherein the periodicity is less than 10 days, preferably less than 7 days.

3. Preparation as claimed in claim 1 or 2, wherein the periodicity amounts to 2-9 days, preferably 2-6 days.

10 4. Preparation as claimed in claims 1-3, wherein the dose of progestogen is substantially constant and the dose of oestrogen oscillates.

5 5. Preparation as claimed in claims 1-3, wherein the dose of progestogen and the dose of estrogen oscillate in such a dose ratio, that blood loss is substantially avoided.

6. Preparation as claimed in claims 1-5, wherein the progestogen comprises

	progesterone	300-900 mg/day
	norethisterone acetate	0.2-5 mg/day
20	medroxyprogesterone acetate	1-5 mg/day
	d-norgestrel	30-150 µgr/day
	desogestrel	30-150 µgr/day
	norgestimate	30-150 µgr/day
	cyproterone acetate	0.2-2 mg/day,
25	gestodene	10-150 µg/day
	3-ketodesogestrel	10-150 µg/day
	drospirenon	0.2-3.0 mg/day

or combinations thereof.

7. Preparation as claimed in claims 1-6, wherein the oestrogen comprises

	aethinylestradiol	5-15 gamma/day
	oestradiol valerianate	1-4 mg/day
	oestradiol	1-2 mg/day
	conjugated oestrogen	0.3-1.25 mg/day

oestriol 1-4 mg/day,
or combinations thereof.

8. Preparation as claimed in claims 1-7, comprising anti-progestogen.

5 9. Preparation as claimed in claim 1-8, comprising anti-estrogen.

10. Preparation as claimed in claims 1-9, containing the progestogen and/or estrogen in an oral, transdermal, parenteral and/or implantable application form.



US005756490A

United States Patent [19][11] **Patent Number:** **5,756,490**

Lachnit et al.

[45] **Date of Patent:** **May 26, 1998**[54] **PHARMACEUTICAL COMBINATION
PREPARATION FOR HORMONAL
CONTRACEPTION**

4,921,843 5/1990 Pasquale 514/170
 5,262,408 11/1993 Berpiak 514/182
 5,280,023 1/1994 Ehrlich et al. 514/177

[75] **Inventors:** Ursula Lachnit; Bernd Düsterberg,
 both of Berlin, Germany; Jürgen
 Spona, Wein, Australia

Primary Examiner—Kimberly Jordan
Attorney, Agent, or Firm—Millen, White, Zelano &
 Branigan, P.C.

[73] **Assignee:** Schering Aktiengesellschaft, Berlin,
 Germany

[57] **ABSTRACT**

[21] **Appl. No.:** 718,401

[22] **PCT Filed:** Mar. 30, 1995

[86] **PCT No.:** PCT/EP95/01190

§ 371 Date: Dec. 16, 1996

§ 102(e) Date: Dec. 16, 1996

[87] **PCT Pub. No.:** WO95/26730

PCT Pub. Date: Oct. 12, 1995

[30] **Foreign Application Priority Data**

Mar. 30, 1994 [DE] Germany 44 11 585.7

[51] **Int. Cl.⁶** A61K 31/56

[52] **U.S. Cl.** 517/170; 514/843

[58] **Field of Search** 514/170, 843

[56] **References Cited****U.S. PATENT DOCUMENTS**

3,502,772 3/1970 Ijzerman 424/239
 3,639,600 2/1972 Hendrix 424/242

A pharmaceutical combination preparation with two hormone components that are manufactured physically separately in a packaging unit and that are intended for time-sequential oral administration, which in each case consist of a number of daily dosage units that are placed physically separately and are individually removable in the packaging unit. As a hormonal active ingredient, a first hormone component contains in combination an estrogen preparation and, in at least a dosage that is sufficient to inhibit ovulation, a gestagen preparation, and as a hormonal active ingredient the second hormone component contains only an estrogen preparation. The first hormone component comprises 23 or 24 daily units and the second hormone component comprises 4 to 10 daily units. The total number of hormone daily units is equal to the total number of days of the desired cycle, but at least 28 days in length. This combination preparation is used for female birth control, and allows for an estrogen content that is as low as possible in each individual dosage unit and also has a low total hormone content per administration cycle, with high contraceptive reliability, low incidence of follicular development, and satisfactory cycle control, with reliable avoidance of intracyclic menstrual bleeding as well as of undesirable side-effects.

13 Claims, No Drawings

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ovulation and thus greater contraceptive reliability, especially in the case of intake errors.

2. The recruitment of the dominant follicle is suppressed as early as in the first cycle by extending the intake of the combination to 23 or 24 days.
3. The intake of 4 to 8 daily estrogen dosage units each in connection with the administration of the 23- or 24-day combination dosage results in considerably improved cycle control and a lower incidence of side-effects, such as headaches, within the framework of the premenstrual syndrome.
4. Other clinical symptoms that are attributable to greatly fluctuating endogenic estrogen levels, such as, for example, breast tenderness, are reduced also clearly 0.015 to 0.025 mg owing to the considerably greater ovarian suppression.

The above-mentioned advantages, especially the suppression of follicular development and the accompanying inhibition of endogenic estrogen production, are more pronounced in the case of the combination preparations according to the invention, which comprise 24 daily units of the first hormone component and thus, as mentioned above, the latter are preferred.

The formulation of an estrogen and a gestagen for the production of a combination preparation according to the invention is carried out completely analogously to the way already known for conventional oral contraceptives with a 21-day intake period of the active ingredients, such as, for example, Femovan® (ethinylestradiol/gestodene) or Microgynon® (ethinylestradiol/levonorgestrel). The formulation of the dosage units that contain only estrogen can also be carried out quite analogously to the way known for already obtained estrogen-containing agents that are intended for oral use, for example, Progynon C®.

A packing that contains a combination preparation according to the invention is also built up analogously to packings for already known oral contraceptives that are on the market, with the difference that, instead of the usual 21 dosage units that contain active components, now 23 or 24 such dosage units and another 4 to 8 dosage units that contain only estrogen are present. As a packaging form for the combination preparation according to the invention, generally a conventional blister pack is used, but other packaging forms that are known for this purpose are also conceivable.

In addition, the invention relates to a process for female contraception in which the above-described combination preparation is administered in the indicated way.

To determine equivalent-action amounts of ethinylestradiol and 17β-estradiol, on the one hand, and various gestagens such as gestodene, levonorgestrel, desogestrel and 3-ketodesogestrel, on the other hand, reference is made to the indications given in EP-A-0 253 607. Other details for determining dose equivalents of various gestagenic active ingredients are found in, for example, "Probleme der Dosisfindung: Sexualhormone [Problems of Dose Finding: Sex Hormones]"; F. Neumann et al., in "Arzneimittelforschung [Pharmaceutical Agent Research]" (Drug Research) 27, 24, 296-318 (1977) as well as in "Aktuelle Entwicklungen in der hormonalen Kontrazeption [Current Developments in Hormonal Contraception]"; H. Kuhl in "Gynäkologie [Gynecologist]" 25: 231-240 (1992).

We claim:

1. Pharmaceutical combination preparation with two hormone components that are manufactured physically separately in a packaging unit and that are intended for time-sequential oral administration, which in each case consist of

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a number of daily dosage units that are placed physically separately and individually removable in the packaging unit, whereby as a hormonal active ingredient a first hormone component contains in combination an estrogen preparation and in at least a dosage that is sufficient to inhibit ovulation a gestagen preparation in either a one-phase or multiple-phase structure and as a hormonal active ingredient the second hormone component contains only an estrogen preparation, whereby the first hormone component comprises 23 or 24 daily units and the second hormone component comprises 4 to 10 daily units, the daily units of the first hormone component do not contain the combination of a biogenic estrogen and a synthetic estrogen, and the total number of hormone daily units is equal to the total number of days of the desired cycle, but at least 28 days in length.

2. Combination preparation according to claim 1, wherein the estrogen of the first hormone component is selected from the group of compounds

17β-estradiol,

ethinylestradiol and

17β-estradiol valerate and the gestagen is selected from the group of compounds

gestodene,

levonorgestrel,

desogestrel,

3-ketodesogestrel,

drospironone,

cyproterone acetate,

norgestimate and

norethisterone and

the estrogen of the second hormone component is selected from the group of compounds

17β-estradiol,

ethinylestradiol and

17β-estradiol valerate.

3. Combination preparation according to claim 2, wherein the estrogen of the first hormone component in each daily dosage unit is contained in a dose of

1.0 to 6.0 mg of 17β-estradiol,

0.015 to 0.025 mg of ethinylestradiol, and

1.0 to 4.0 mg of 17β-estradiol valerate

and the gestagen in each daily dosage unit is contained in a dose of

0.05 to 0.075 mg of gestodene,

0.05 to 0.125 mg of levonorgestrel,

0.06 to 0.15 mg of desogestrel,

0.06 to 0.15 mg of 3-ketodesogestrel,

1.0 to 3.0 mg of drospironone,

1.0 to 2.0 mg of cyproterone acetate,

0.2 mg to 0.3 mg of norgestimate and

0.35 to 0.75 mg of norethisterone.

4. The combination preparation of claim 3, wherein the second hormone component contains, in each daily dosage unit, an amount of:

1.0 to 6.0 mg of 17β-estradiol,

0.002 to 0.04 mg of ethinylestradiol, and

1.0 to 4.0 mg of 17β-estradiol valerate.

5. Combination preparation according to claim 2, wherein the second hormone component contains, in each daily dosage unit, an amount of

1.0 to 6.0 mg of 17β-estradiol,

0.002 to 0.04 mg of ethinylestradiol, and

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1.0 to 4.0 mg of 17 β -estradiol valerate.

6. Combination preparation according to claim 5, wherein the second hormone component in each daily dosage unit contains ethinylestradiol in an amount of 0.01 to 0.025 mg.

7. The combination preparation of claim 2, wherein the second hormone component contains, in each daily dosage unit, an amount of:

1.0 to 3.0 mg of 17 β -estradiol.

0.01 to 0.025 mg of ethinylestradiol, and

1.0 to 4.0 mg of 17 β -estradiol valerate.

8. Combination preparation according to claim 1, wherein the total number of hormone daily units is 28.

9. The combination preparation according to claim 1, wherein the first hormone component has 24 daily units and the second hormone component has 4-8 daily units.

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10. The combination preparation according to claim 1, wherein the first hormone component has 24 daily units and the second hormone component has 4 daily units.

11. The combination preparation of claim 1, wherein the estrogen in both the first and second hormone components is selected from ethinylestradiol or 17 β -estradiol.

12. The combination preparation of claim 1, wherein the gestagen in the first hormone component is selected from gestodene or levonorgestel.

13. The combination preparation of claim 1, wherein the first hormone component has three phases of daily units wherein the estrogen and/or the gestagen amounts differ between phases.

* * * * *

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WELTORGANISATION FÜR GEISTIGES EIGENTUM
Internationales Büro
INTERNATIONALE ANMELDUNG VERÖFFENTLICHT NACH DEM VERTRAG ÜBER DIE
INTERNATIONALE ZUSAMMENARBEIT AUF DEM GEBIET DES PATENTWESENS (PCT)

(51) Internationale Patentklassifikation ⁶ : A61K 31/57, 31/565	A1	(11) Internationale Veröffentlichungsnummer: WO 95/26730 (43) Internationales Veröffentlichungsdatum: 12. Oktober 1995 (12.10.95)
(21) Internationales Aktenzeichen: PCT/EP95/01190 (22) Internationales Anmeldedatum: 30. März 1995 (30.03.95) (30) Prioritätsdaten: P 44 11 585.7 30. März 1994 (30.03.94) DE (71) Anmelder (für alle Bestimmungsstaaten ausser US): SCHERING AKTIENGESELLSCHAFT [DE/DE]; Müllerstrasse 178, D-13353 Berlin (DE). (72) Erfinder; und (75) Erfinder/Anmelder (nur für US): LACHNIT, Ursula [DE/DE]; Livländische Strasse 17, D-10715 Berlin (DE). DÜSTERBERG, Bernd [DE/DE]; Spirdingseestrasse 27, D-12307 Berlin (DE). SPONA, Jürgen [AT/AT]; Hameau Strasse 19/9, A-1190 Wien (AT).	(81) Bestimmungsstaaten: AU, BG, BR, CA, CN, CZ, EE, FI, HU, JP, KR, LT, LV, MX, NO, NZ, PL, RO, RU, SI, SK, UA, US, europäisches Patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE). Veröffentlicht <i>Mit internationalem Recherchenbericht. Vor Ablauf der für Änderungen der Ansprüche zugelassenen Frist. Veröffentlichung wird wiederholt falls Änderungen eintreffen.</i>	
(54) Title: COMBINED HORMONAL CONTRACEPTION PHARMACEUTICAL PREPARATION (54) Bezeichnung: PHARMAZEUTISCHES KOMBINATIONSPRÄPARAT ZUR HORMONALEN KONTRAZEPTION (57) Abstract A combined pharmaceutical preparation has two hormonal components for sequential oral administration separately conditioned in a packaging unit that consist of a number of daily dosing units separately conditioned in and individually removable from the packaging unit. The first hormonal component contains as active hormonal substance a dose of oestrogen combined with a dose of a gestagen preparation that is at least sufficient to inhibit ovulation. The second hormonal component contains as active hormonal substance only an oestrogen preparation. The first hormonal component consists of 23 or 24 daily units and the second hormonal component of 4 to 10 daily units. The total number of hormonal daily units equals the total number of days in the desired cycle, but at least 28 days. This combined female contraception preparation allows the oestrogen content in each individual dosing unit to be reduced as much as possible and also has a low total hormonal content per administration cycle, while ensuring a high contraceptive dependability, a low incidence of follicle development, a flawless cycle control and a reliable prevention of spotting and other undesirable side effects. (57) Zusammenfassung Die vorliegende Erfindung beschreibt ein pharmazeutisches Kombinationspräparat mit zwei in einer Verpackungseinheit räumlich getrennt konfektionierten, zur zeitlich sequentiellen oralen Verabreichung bestimmten Hormonkomponenten, die jeweils aus einer Anzahl räumlich getrennt und einzeln entnehmbar in der Verpackungseinheit untergebrachten täglichen Dosierungseinheiten bestehen, wobei eine erste der Hormonkomponenten als hormonellen Wirkstoff in Kombination ein Estrogen- und in mindestens zur Ovulationshemmung ausreichender Dosierung ein Gestagenpräparat und die zweite Hormonkomponente als hormonellen Wirkstoff lediglich ein Estrogenpräparat enthält, wobei die erste Hormonkomponente 23 oder 24 und die zweite Hormonkomponente 4 bis 10 Tageseinheiten umfaßt und die Gesamtzahl der Hormontageseinheiten gleich der Gesamtzahl der Tage des gewünschten, mindestens aber 28 Tage langen, Zyklus ist. Dieses Kombinationspräparat, das zur weiblichen Fertilitätskontrolle dient, gestattet einen möglichst niedrigen Estrogengehalt in jeder einzelnen Dosierungseinheit und weist auch einen niedrigen Gesamthormongehalt pro Verabreichungszyklus auf, bei hoher kontrazeptiver Sicherheit, geringer Inzidenz an Follikelentwicklung, einwandfreier Zykluskontrolle unter zuverlässiger Vermeidung von Zwischenblutungen sowie unerwünschter Nebenwirkungen.		

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International Bureau



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶: A61K 31/565, 31/57	A1	(11) International Publication Number: WO 98/04267 (43) International Publication Date: 5 February 1998 (05.02.98)
(21) International Application Number: PCT/US97/12789 (22) International Filing Date: 23 July 1997 (23.07.97) (30) Priority Data: 08/687,855 26 July 1996 (26.07.96) US (71) Applicant: AMERICAN HOME PRODUCTS CORPORATION (US/US); Five Giralda Farms, Madison, NJ 07940-0874 (US). (72) Inventor: GAST, Michael, Jay; 230 Country Lane, Phoenixville, PA 19460 (US). (74) Agents: ALICE, Ronald, W.; American Home Products Corporation, Patent Law Dept. - 2B, One Campus Drive, Parsippany, NJ 07054 (US) et al.	(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, ARIPO patent (GH, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i> <i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>	
(54) Title: PROGESTIN/ESTROGEN ORAL CONTRACEPTIVE (57) Abstract This invention provides a method of contraception which comprises administering to a female of child-bearing age a combination of a progestin at a daily dosage of 40-500 µg trimegestone, 250 µg - 4 mg dienogest, or 250 µg - 4 mg drospirenone; and an estrogen at a daily dosage equivalent in estrogenic activity to 10-20 µg ethinyl estradiol for 23-25 days beginning on day 1 of the menstrual cycle; wherein the same dosage of the progestin and estrogen combination is administered in each of the 23-25 days, followed by the administration of an estrogen at a daily dosage equivalent in estrogenic activity to 5-15 µg ethinyl estradiol for 3-5 days; wherein the same dosage of the estrogen is administered in each of the 3-5 days; such that the number of days of administration of the progestin and estrogen combination plus the number of days of administration of estrogen is equal to 28 per menstrual cycle.		

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WHAT IS CLAIMED IS:

1. A method of contraception which comprises administering to a female of child bearing age a combination of a progestin at a daily dosage selected from the group consisting of 40-500 µg trimegestone, 250 µg - 4 mg dienogest, and 250 µg - 4 mg drospirenone, and an estrogen at a daily dosage equivalent in estrogenic activity to 10-20 µg ethinyl estradiol for 23-25 days beginning on day 1 of the menstrual cycle; wherein the same dosage of the progestin and estrogen combination is administered in each of the 23-25 days, followed by the administration of an estrogen at a daily dosage equivalent in estrogenic activity to 5-15 µg ethinyl estradiol for 3-5 days; wherein the same dosage of the estrogen is administered in each of the 3-5 days; such that the number of days of administration of the progestin and estrogen combination plus the number of days of administration of estrogen is equal to 28 per menstrual cycle.
2. The method according to claim 1, wherein the estrogen is selected from the group consisting of ethinyl estradiol; 17β-estradiol; conjugated estrogens, USP; estrone or a salt thereof; and mestranol.
3. The method according to claim 2, wherein the estrogen is ethinyl estradiol.
4. The method according to claim 3, wherein the progestin/estrogen combination is administered for 24 days per menstrual cycle beginning on day 1 of the menstrual cycle.
5. The method according to claim 4 wherein the daily dosage of ethinyl estradiol during the 24 day administration period is 15 µg.
6. The method according to claim 5, wherein the progestin is trimegestone.
7. The method according to claim 6, wherein the daily dosage of trimegestone is 250 µg.
8. The method according to claim 6, wherein the daily dosage of trimegestone is 125 µg.

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9. The method according to claim 6, wherein the daily dosage of
trimegestone is 75 μ g.
10. The method according to claim 6, wherein the daily dosage of
5 trimegestone is 50 μ g.
11. The method according to claim 6, wherein the daily dosage of
trimegestone is 40 μ g.
- 10 12. The method according to claim 5, wherein the progestin is dienogest.
13. The method according to claim 11, wherein the daily dosage of dienogest
is 2 mg.
- 15 14. The method according to claim 11, wherein the daily dosage of dienogest
is 1 mg.
15. The method according to claim 11, wherein the daily dosage of dienogest
is 800 μ g.
- 20 16. The method according to claim 11, wherein the daily dosage of dienogest
is 750 μ g.
17. The method according to claim 11, wherein the daily dosage of dienogest
25 is 500 μ g.
18. The method according to claim 5, wherein the progestin is drospirenone.
19. The method according to claim 18, wherein the daily dosage of
30 drospirenone is 3 mg.
20. The method according to claim 18, wherein the daily dosage of
drospirenone is 2 mg.
- 35 21. The method according to claim 18, wherein the daily dosage of
drospirenone is 1 mg.

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22. The method according to claim 18, wherein the daily dosage of drospirenone is 500 μ g.

5 23. A contraceptive kit adapted for daily oral administration which comprises
28 separate dosage units, 23-25 separate dosage units, said dosage units each
containing a combination of a progestin at a dosage selected from the group
consisting of 40-500 μ g trimegestone, 250 μ g - 4 mg dienogest, and 250 μ g - 4
10 mg drospirenone, and an estrogen at a dosage equivalent in estrogenic activity to
10-20 μ g ethinyl estradiol, wherein each of the dosage units contains the same
dosage of progestin and estrogen; and 3-5 of said dosage units containing an
estrogen at a daily dosage equivalent in estrogenic activity to 5-15 μ g ethinyl
estradiol, wherein each of the 3-5 dosage units contains the same dosage of
estrogen.

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24. The contraceptive kit according to claim 23, wherein the estrogen is
selected from the group consisting of ethinyl estradiol; 17 β -estradiol; conjugated
estrogens, USP; estrone or a salt thereof; and mestranol.

20 25. The contraceptive kit according to claim 24, wherein the estrogen is
ethinyl estradiol.

25 26. The contraceptive kit according to claim 25, wherein 24 of the 28 dosage
units contain the progestin and estrogen combination, and 4 of the dosage units
are progestin free.



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INTERNATIONALE ANMELDUNG VERÖFFENTLICHT NACH DEM VERTRAG ÜBER DIE
INTERNATIONALE ZUSAMMENARBEIT AUF DEM GEBIET DES PATENTWESENS (PCT)

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(51) Internationale Patentklassifikation ⁶: A61K	A2	(11) Internationale Veröffentlichungsnummer: WO 98/27929 (43) Internationales Veröffentlichungsdatum: 2. Juli 1998 (02.07.98)
(21) Internationales Aktenzeichen: PCT/DE97/03032 (22) Internationales Anmeldedatum: 22. Dezember 1997 (22.12.97) (30) Prioritätsdaten: 196 54 609.5 20. Dezember 1996 (20.12.96) DE (71) Anmelder (für alle Bestimmungsstaaten ausser US): SCHERING AKTIENGESELLSCHAFT [DE/DE]; Müllerstrasse 178, D-13353 Berlin (DE). (72) Erfinder; und (75) Erfinder/Anmelder (nur für US): NASHED, Norman [FR/DE]; Margeritenweg 24, D-14532 Stahnsdorf (DE).	(81) Bestimmungsstaaten: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO Patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), eurasisches Patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), europäisches Patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI Patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Veröffentlicht <i>Ohne internationalen Recherchenbericht und erneut zu veröffentlichen nach Erhalt des Berichts.</i>	
(54) Title: THERAPEUTIC GESTAGENS FOR THE TREATMENT OF PREMENSTRUAL DYSPHORIC DISORDER (54) Bezeichnung: THERAPEUTISCHE GESTAGENE ZUR BEHANDLUNG VON PREMENSTRUAL DYSPHORIC DISORDER (57) Abstract The invention relates to the use of a therapeutic gestagen (for example drospirenon, cyproteron acetate, dienogest) for the production of a medicament for the treatment of Premenstrual Dysphoric Disorders (PMDD), possibly in combination with a natural or synthetic oestrogen (for example estradiol or ethinyl estradiol). (57) Zusammenfassung Die vorliegende Erfindung beschreibt die Verwendung eines therapeutischen Gestagens (z.B. Drospirenon, Cyproteronacetat, Dienogest) zur Herstellung eines Arzneimittels zur Behandlung von Premenstrual Dysphoric Disorders (PMDD), gegebenenfalls in Kombination mit einem natürlichen oder synthetischen Estrogen (z.B. Estradiol bzw. Ethinylestradiol).		

Patentansprüche

1. Verwendung eines therapeutischen Gestagens zur Herstellung eines Arzneimittels zur Behandlung von Premenstrual Dysphoric Disorders (PMDD).
2. Verwendung von Drospirenon, Cyproteronacetat, Dienogest gemäß Anspruch 1.
3. Verwendung gemäß Anspruch 1 gemeinsam mit einem Estrogen.
4. Verwendung gemäß Anspruch 3 gemeinsam mit einem synthetischen Estrogen.
5. Verwendung gemäß Anspruch 4 gemeinsam mit Ethinylestradiol.
6. Verwendung gemäß Anspruch 3 gemeinsam mit einem Estrogensulfamat.
7. Verwendung gemäß Anspruch 3 gemeinsam mit einem natürlichen Estrogen.
8. Verwendung gemäß Anspruch 7 gemeinsam mit Estradiol, Estradiolvalerat oder einem anderen Estradiolester.
9. Verwendung gemäß Anspruch 1 ausschließlich während der lutealen Phase des weiblichen Menstruationscyclus.
10. Verwendung gemäß Anspruch 9 von Tag 10 bis 28 des Menstruationscyclus.
11. Verwendung von Drospirenon gemäß Anspruch 1 in einer Menge von 0,5 mg bis weniger als 5 mg täglich.
12. Verwendung von Ethinylestradiol gemäß Anspruch 5 in einer Menge von 0,010 bis 0,05 mg täglich.
13. Verwendung von Estradiol gemäß Anspruch 8 in einer Menge von 1,0 bis 3,0 mg täglich.



US005583129A

United States Patent [19]
Spona et al.

[11] **Patent Number:** **5,583,129**
[45] **Date of Patent:** **Dec. 10, 1996**

[54] **COMPOSITION FOR CONTRACEPTION**

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A. R. Genazzani, et al. (Ed), Progress in Gynecology and
Obstetrics, "Multicenter Clinical Trial on the new Oral
Contraceptive 20 µg Ethinylestradiol", Chapter 1, pp.
747-756, (1990).

Primary Examiner—Phyllis G. Spivack
Attorney, Agent, or Firm—Millen, White, Zelano, & Branigan, P.C.

[21] **Appl. No.:** **268,996**

[22] **Filed:** **Jun. 30, 1994**

[30] **Foreign Application Priority Data**

Dec. 22, 1993 [DE] Germany 43 44 462.8

[51] **Int. Cl.⁶** **A61K 31/56; A61K 31/57;**
A61K 31/585

[52] **U.S. Cl.** **514/178; 514/173; 514/182**

[58] **Field of Search** **514/178, 182,**
514/173

[56] **References Cited**

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Plus 20 µg Ethinylestradiol, a Triphasic Combination Con-
taining Levonorgestrel and a Monophasic Combination
Containing gestrodene on Coagulatory Factors", vol. 43,
No. 1, pp. 23-30 (Jan. 1991).

[57] **ABSTRACT**

This invention relates to a method of inducing contraception
comprising administering an estrogen selected from

2.0 to 6.0 mg of 17β-estradiol and
0.015 to 0.020 mg of ethinylestradiol;
and a gestagen selected from
0.05 to 0.075 mg of gestodene,
0.075 to 0.125 mg of levonorgestrel,
0.06 to 0.15 mg of desogestrel,
0.06 to 0.15 mg of 3-ketodesogestrel,
0.1 to 0.3 mg of drospirenone,
0.1 to 0.2 mg of cyproterone acetate,
0.2 to 0.3 mg of norgestimate and
>0.35 to 0.75 mg of norethisterone

for a female of reproductive age, who has not yet reached
premenopause, by administration for 23 or 24 days, begin-
ning on day one of the menstrual cycle, followed by 5 or 4
pill-free or sugar pill days, during a total of 28 days in the
administration cycle.

8 Claims, 1 Drawing Sheet

the 23-day preparation, especially in the case of previous intake errors. The danger of "breakthrough ovulations" is smaller.

2. The occurrence of large follicles of more than a 30 mm diameter is extremely rare. The development of ovarian cysts is improbable with the 23-day preparation in comparison to the 21-day preparation.
3. The recruitment of dominant follicles is suppressed in the shortened intake-free pause.
4. The endogenous 17 β -estradiol levels are suppressed easily controllably in the case of the majority of the users of the 23-day preparation. Clinical symptoms such as breast tenderness, premenstrual syndrome and menstrual disorders, which can be attributed to increased and greatly fluctuating estrogen levels, are observed with the 23-day preparation with clearly lower frequency.

In summary, an intake, extended by two (or three) days, of preparations containing 20 μ g of ethinylestradiol in each daily dosage unit can produce the above-mentioned advantages, without the daily dose having to be raised to the previously largely used level of 30 μ g of ethinylestradiol.

The formulation of an estrogen and gestagen for the use according to the invention or for a combination preparation according to the invention takes place completely analogously as it is already known for usual oral contraceptives with 21-day intake period of the active ingredients, such as, for example, Femovan® (ethinylestradiol/gestodene) or Microgynon® (ethinylestradiol/levonorgestrel).

A pack containing a combination preparation according to the invention is also designed analogously to packs for already known oral contraceptives on the market with the variation that instead of the usual 21 dosage units containing the active components, now 23 or 24 such dosage units and 5 or 4 sugar pills are present or else contain other suitable indications that 5 or 4 days are to be bridged over until continuation of the intake of active ingredient-containing dosage units.

Moreover, reference is made to the statements made in EP-A 0 253 607, especially also to the statements there for determination of equivalent amounts of ethinylestradiol and 17 β -estradiol, on the one hand, and various gestagens, such as levonorgestrel, desogestrel, 3-ketodesogestrel and gestodene, on the other hand.

For further details for the determination of dose equivalents of various gestagenic active ingredients, reference is made to "Probleme der Dosisfindung: Sexualhormone" [Problems of Dose-Finding: Sex Hormones]; F. Neumann et al. in "Arzneimittelforschung" (Pharmaceutical Agent Research) 27, 2a, 296-318 (1977), as well as to "Aktuelle Entwicklungen in der hormonalen Kontrazeption" [Current Developments in Hormonal Contraception]; H. Kuhl in Gynäkologe" [Gynecologist] 25: 231-240 (1992).

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1: Area with the 17 β -estradiol level in groups of 30 females, who are treated with an oral contraceptive (75 μ g of gestodene +20 μ g of ethinylestradiol) in 21- or 23-day administration interval over three cycles.

FIG. 2: Number of females in %, who showed follicular developments (>13 mm diameter) with 21- or 23-day treatment with an oral contraceptive (75 μ g of gestodene +20 μ g of ethinylestradiol).

We claim:

1. A method of inducing contraception in a female of reproductive age who has not yet reached premenopause, comprising administering to said female a composition comprising an estrogen selected from

- 2.0 to 6.0 mg of 17 β -estradiol and
- 0.015 to 0.020 mg of ethinylestradiol;
- and a gestagen selected from
- 0.05 to 0.075 mg of gestodene,
- 0.075 to 0.125 mg of levonorgestrel,
- 0.06 to 0.15 mg of desogestrel,
- 0.06 to 0.15 mg of 3-ketodesogestrel,
- 0.1 to 0.3 mg of drospirenone,
- 0.1 to 0.2 mg of cyproterone acetate,
- 0.2 to 0.3 mg of norgestimate and
- >0.35 to 0.75 mg of norethisterone;

wherein the composition is administered for 23 or 24 days, beginning on day one of the menstrual cycle, followed by 5 or 4 pill-free or sugar pill days, during a total of 28 days in the administration cycle.

2. A method according to claim 1, whereby the estrogen is ethinylestradiol.

3. A method according to claim 1, whereby the estrogen is 17 β -estradiol.

4. A method according to claim 1, whereby the gestagen is gestodene.

5. A method according to claim 1, whereby the gestagen is levonorgestrel.

6. A method according to claim 1, whereby the gestagen is cyproterone acetate or drospirenone.

7. A method according to claim 1, whereby the composition comprises an estrogen selected from

- >2.0 to 6.0 mg of 17 β -estradiol and
- 0.020 mg of ethinylestradiol;
- and a gestagen selected from
- >0.06 to 0.075 mg of gestodene,
- >0.100 to 0.125 mg of levonorgestrel,
- >0.10 to 0.15 mg of desogestrel,
- >0.10 to 0.15 mg of 3-ketodesogestrel,
- 0.25 to 0.30 mg of drospirenone,
- 0.1 to 0.2 mg of cyproterone acetate,
- 0.2 to 0.3 mg of norgestimate and
- 0.50 to 0.75 mg of norethisterone.

8. A method according to claim 1, whereby the estrogen is present in a dose of 20 μ g of ethinylestradiol or an equivalent dose of 17 β -estradiol and the gestagen is present in a dose of 75 μ g of gestodene or an equivalent dose of levonorgestrel, cyproterone acetate or drospirenone.

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INTERNATIONALE ANMELDUNG VERÖFFENTLICHT NACH DEM VERTRAG ÜBER DIE
INTERNATIONALE ZUSAMMENARBEIT AUF DEM GEBIET DES PATENTWESENS (PCT)

(51) Internationale Patentklassifikation ⁶ : A61K 31/57, 31/565		A1	(11) Internationale Veröffentlichungsnummer: WO 95/17194
			(43) Internationales Veröffentlichungsdatum: 29. Juni 1995 (29.06.95)
(21) Internationales Aktenzeichen: PCT/EP94/04274		(81) Bestimmungsstaaten: CA, CN, CZ, HU, JP, KR, LT, LV, NO, NZ, PL, RU, SI, SK, UA, europäisches Patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).	
(22) Internationales Anmeldedatum: 22. December 1994 (22.12.94)			
(30) Prioritätsdaten: P 43 44 462.8 22. December 1993 (22.12.93) DE		Veröffentlicht Mit internationalem Recherchenbericht. Vor Ablauf der für Änderungen der Ansprüche zugelassenen Frist. Veröffentlichung wird wiederholt falls Änderungen eintreffen.	
(71) Anmelder: SCHERING AKTIENGESELLSCHAFT [DE/DE]; Müllerstrasse 178, D-13353 Berlin (DE).			
(72) Erfinder: SPONA, Jürgen; Billrothstrasse 78, A-1190 Wien (AT). DÜSTERBERG, Bernd; Spirdingseestrasse 27, D-12307 Berlin (DE). LÜDICKE, Frank; Hopital Cantonal Universitaire, Clinic of Sterility and Gynaecologic Endocrinology, 32bis, boulevard de la Cluse, CH-1211 Geneva 4 (CH). FEICHTINGER, W.; Trauttmansdorffgasse 3a, A-1130 Wien (AT). ELSTEIN, Max; University of Manchester, Dept. of Obstetrics & Gynecology, Nell Lane, West Didsbury, Manchester M20 8LR (GB).			
(54) Title: COMPOSITION FOR A CONTRACEPTIVE COMPRISING AN ESTROGEN AND A GESTAGEN			
(54) Bezeichnung: ZUSAMMENSETZUNG FÜR DIE EMPFÄNGNISVERHÜTUNG UMFASSEND EIN ESTROGEN UND EIN GESTAGEN			
(57) Abstract			
The invention concerns the use of a composition comprising an estrogen selected from 2.0-6.0 mg 17β-estradiol and 0.015-0.020 mg ethinyl estradiol and a gestagen selected from 0.05-0.075 mg gestodene, 0.075-0.125 mg levonorgestrel, 0.06-0.15 mg desogestrel, 0.06-0.15 mg 3-ketodesogestrel, 0.1-0.3 mg drospirenone, 0.1-0.2 mg cyproterone acetate, 0.2-0.3 mg norgestimate and >0.35-0.75 mg norethisterone, used to produce a dosage formulation for contraception for a woman of child-bearing age who has not reached premenopause by administering the dosage formulation during 23 or 24 days, beginning on day one of the menstrual cycle, followed by 5 or 4 pill-free or placebo days, over a complete 28-day cycle of administration, wherein the individual dosage formulations contain a constant amount of estrogen/gestagen throughout the 23 or 24 days. The invention also pertains to a combination oral contraceptive based on this composition.			
(57) Zusammenfassung			
Die vorliegende Erfindung betrifft die Verwendung einer Zusammensetzung umfassend ein Estrogen ausgewählt aus 2,0 bis 6,0 mg 17β-Estradiol und 0,015 bis 0,020 mg Ethinylestradiol; und ein Gestagen ausgewählt aus 0,05 bis 0,075 mg Gestoden, 0,075 bis 0,125 mg Levonorgestrel, 0,06 bis 0,15 mg Desogestrel, 0,06 bis 0,15 mg 3-Ketodesogestrel, 0,1 bis 0,3 mg Drospirenon, 0,1 bis 0,2 mg Cyproteronacetat, 0,2 bis 0,3 mg Norgestimat und >0,35 bis 0,75 mg Norethisteron zur Herstellung einer Dosierungsform für die Empfängnisverhütung für eine Frau im fertilen Alter, die die Prämenopause noch nicht erreicht hat, durch Verabreichung der Dosierungsform während 23 oder 24 Tagen, beginnend am Tag eins des Menstruationszyklus, gefolgt von 5 oder 4 pillenfreien oder Blindpillentagen, während insgesamt 28 Tagen im Verabreichungszyklus, wobei die einzelnen Dosierungsformen über alle 23 oder 24 Tage eine konstante Estrogen/Gestagenmenge enthalten, sowie ein monophasisches Kombinationsprodukt für die orale Kontrazeption auf der Basis dieser Zusammensetzung.			

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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

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(21) International Application Number: PCT/US97/12795 (22) International Filing Date: 23 July 1997 (23.07.97) (30) Priority Data: 08/686,790 26 July 1996 (26.07.96) US (71) Applicant: AMERICAN HOME PRODUCTS CORPORATION [US/US]: Five Giralda Farms, Madison, NJ 07940-0874 (US). (72) Inventor: GAST, Michael, Jay: 230 Country Lane, Phoenixville, PA 19460 (US). (74) Agents: ALICE, Ronald, W.; American Home Products Corporation, Patent Law Dept. - 2B, One Campus Drive, Parsippany, NJ 07054 (US) et al.	(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, ARIPO patent (GH, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>	
(54) Title: MONOPHASIC CONTRACEPTIVE METHOD AND KIT COMPRISING A COMBINATION OF A PROGESTIN AND ESTROGEN (57) Abstract This invention provides a method of contraception which comprises administering to a female of child bearing age for a combination of a progestin at a daily dosage of 40-500 µg trimegestone, 250 µg - 4 mg dienogest, or 250 µg - 4 mg drospirenone, and an estrogen at a daily dosage equivalent in estrogenic activity to 10-20 µg ethinyl estradiol for 23-25 days beginning on day 1 of the menstrual cycle, and wherein the same dosage of the progestin and estrogen combination is administered in each of the 23-25 days.		

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WHAT IS CLAIMED IS:

1. A method of contraception which comprises administering to a female of child bearing age a combination of a progestin at a daily dosage selected from the group consisting of 40-500 μg trimegestone, 250 μg - 4 mg dienogest, and 250 μg - 4 mg drospirenone, and an estrogen at a daily dosage equivalent in estrogenic activity to 10-20 μg ethinyl estradiol for 23-25 days beginning on day 1 of the menstrual cycle, and wherein the same dosage of the progestin and estrogen combination is administered in each of the 23-25 days.
2. The method according to claim 1, wherein the estrogen is selected from the group consisting of ethinyl estradiol; 17 β -estradiol; conjugated estrogens, USP; estrone or a salt thereof; and mestranol.
3. The method according to claim 2, wherein the estrogen is ethinyl estradiol.
4. The method according to claim 3, wherein the progestin/estrogen combination is administered for 24 days per menstrual cycle beginning on day 1 of the menstrual cycle.
5. The method according to claim 4 wherein the daily dosage of ethinyl estradiol is 15 μg .
6. The method according to claim 5, wherein the progestin is trimegestone.
7. The method according to claim 6, wherein the daily dosage of trimegestone is 250 μg .
8. The method according to claim 6, wherein the daily dosage of trimegestone is 125 μg .
9. The method according to claim 6, wherein the daily dosage of trimegestone is 75 μg .
10. The method according to claim 6, wherein the daily dosage of trimegestone is 50 μg .

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11. The method according to claim 6, wherein the daily dosage of
trimegestone is 40 µg.
- 5 12. The method according to claim 5, wherein the progestin is dienogest.
13. The method according to claim 11, wherein the daily dosage of dienogest
is 2 mg.
- 10 14. The method according to claim 11, wherein the daily dosage of dienogest
is 1 mg.
- 15 15. The method according to claim 11, wherein the daily dosage of dienogest
is 800 µg.
16. The method according to claim 11, wherein the daily dosage of dienogest
is 750 µg.
17. The method according to claim 11, wherein the daily dosage of dienogest
20 is 500 µg.
18. The method according to claim 5, wherein the progestin is drospirenone.
19. The method according to claim 18, wherein the daily dosage of
25 drospirenone is 3 mg.
20. The method according to claim 18, wherein the daily dosage of
drospirenone is 2 mg.
- 30 21. The method according to claim 18, wherein the daily dosage of
drospirenone is 1 mg.
22. The method according to claim 18, wherein the daily dosage of
drospirenone is 500 µg.

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23. The method according to claim 1, which further comprises administering a non-contraceptive placebo for 3-5 consecutive days beginning on the day immediately following the last day of administration of the progestin/estrogen combination, such that the total days of administration of the progestin/estrogen combination plus the non-contraceptive placebo equals 28 days.

24. The method according to claim 23 in which the non-contraceptive placebo contains an iron supplement.

25. A contraceptive kit adapted for oral administration which comprises 23-25 separate dosage units, said dosage units each containing a combination of a progestin at a dosage selected from the group consisting of 40-500 µg trimegestone, 250 µg - 4 mg dienogest, and 250 µg - 4 mg drospirenone, and an estrogen at a dosage equivalent in estrogenic activity to 10-20 µg ethinyl estradiol, wherein each of the dosage units contains the same dosage of progestin and estrogen.

26. The contraceptive kit according to claim 25, wherein the estrogen is selected from the group consisting of ethinyl estradiol; 17β-estradiol; conjugated estrogens, USP; estrone or a salt thereof; and mestranol.

27. The contraceptive kit according to claim 26, wherein the estrogen is ethinyl estradiol.

28. The contraceptive kit according to claim 27 in which the number of dosage units is 24.

29. A contraceptive kit adapted for oral administration which comprises 28 dosage separate units, 23-25 of said dosage units each containing each containing a combination of a progestin at a dosage selected from the group consisting of 40-500 µg trimegestone, 250 µg - 4 mg dienogest, and 250 µg mg drospirenone, and an estrogen at a dosage equivalent in estrogenic activity to 10-20 µg ethinyl estradiol, wherein each of the dosage units contains the same dosage of progestin and estrogen; and 3-5 of said dosage units each containing a non-contraceptive placebo; wherein each of the 23-25 dosage units contains the same dosage of progestin and estrogen.

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INTERNATIONALE ANMELDUNG VERÖFFENTLICHT NACH DEM VERTRAG ÜBER DIE
INTERNATIONALE ZUSAMMENARBEIT AUF DEM GEBIET DES PATENTWESENS (PCT)

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(21) Internationales Aktenzeichen: PCT/DE96/01192 (22) Internationales Anmeldedatum: 27. Juni 1996 (27.06.96) (30) Prioritätsdaten: 195 25 017.6 28. Juni 1995 (28.06.95) DE (71) Anmelder (für alle Bestimmungsstaaten ausser US): SCHERING AKTIENGESELLSCHAFT [DE/DE]; Müllerstrasse 178, D-13353 Berlin (DE). (72) Erfinder; und (75) Erfinder/Anmelder (nur für US): SCHMIDT-GOLLWITZER, Karin [DE/DE]; Palmzeile 10 B, D-14129 Berlin (DE). KLEMMANN, Walter [DE/DE]; Kastanienallee 21, D-14052 Berlin (DE).	(81) Bestimmungsstaaten: AU, BR, CA, CN, CZ, FI, HU, IL, JP, KR, MX, NO, NZ, PL, RU, SK, UA, US, europäisches Patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE). Veröffentlicht <i>Mit internationalem Recherchenbericht. Vor Ablauf der für Änderungen der Ansprüche zugelassenen Frist. Veröffentlichung wird wiederholt falls Änderungen eintreffen.</i>	
(54) Title: PHARMACEUTICAL COMBINED PREPARATION, KIT AND METHOD FOR HORMONAL CONTRACEPTION (54) Bezeichnung: PHARMAZEUTISCHES KOMBINATIONSPRÄPARAT, KIT UND METHODE ZUR HORMONALEN KONTRAZEPTION (57) Abstract The invention concerns a two-stage pharmaceutical combined preparation for hormonal contraception containing at least 30 daily doses. The hormonal active substance in the first stage is a combination of an oestrogen preparation and a gestagen preparation in single-stage form in a quantity at least sufficient to inhibit ovulation. In the second stage, the hormonal active substance is an oestrogen preparation only. The first stage comprises not less than 25 and not more than 77 daily discrete or continuous doses and the second stage comprises 5, 6 or 7 daily discrete or continuous doses, the total number of daily units being equal to the total number of days of the desired cycle, i.e. at least thirty and not more than eighty four. This combined preparation can be made available in, for example, monthly packaging and is used for controlling female fertility. It reduces to a minimum the oestrogen content in each individual dose and also has a low overall hormone content for the dosing cycle, while ensuring a high contraceptive reliability, low incidence of follicle development, excellent cycle control and reliable prevention of intercycle menstrual bleeding and unwanted side effects. (57) Zusammenfassung Die vorliegende Erfindung beschreibt ein zweistufiges pharmazeutisches Kombinationspräparat zur hormonalen Kontrazeption enthaltend mindestens 30 tägliche Dosierungseinheiten, welches in seiner ersten Stufe als hormonellen Wirkstoff in Kombination ein Estrogen- und in mindestens zur Ovulationshemmung ausreichender Dosierung ein Gestagenpräparat in einstufiger Ausbildung und in der zweiten Stufe als hormonellen Wirkstoff lediglich ein Estrogenpräparat enthält, wobei die erste Stufe mindestens 25 und maximal 77 diskrete tägliche oder kontinuierliche Dosierungseinheiten und die zweite Stufe 5, 6 oder 7 tägliche diskrete oder kontinuierliche Dosierungseinheiten umfaßt und die Gesamtzahl der Tageseinheiten gleich der Gesamtzahl der Tage des gewünschten, mindestens 30 und maximal 84 Tage langen, Zyklus ist. Dieses beispielsweise als Monatspackung vorliegende Kombinationspräparat, das zur weiblichen Fertilitätskontrolle dient, gestattet einen möglichst niedrigen Estrogengehalt in jeder einzelnen Dosierungseinheit und weist auch einen niedrigen Gesamthormongehalt pro Verabreichungszyklus auf, bei hoher kontrazeptiver Sicherheit, geringer Inzidenz an Follikelentwicklung, einwandfreier Zykluskontrolle unter zuverlässiger Vermeidung von Zwischenblutungen sowie unerwünschter Nebenwirkungen.		

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322**Patentansprüche**

1. Zweistufiges pharmazeutisches Kombinationspräparat zur hormonalen
5 Kontrazeption enthaltend mindestens 30 tägliche Dosierungseinheiten, welches in
seiner ersten Stufe als hormonellen Wirkstoff in Kombination ein Estrogen- und in
mindestens zur Ovulationshemmung ausreichender Dosierung ein Gestagenpräparat
in einstufiger Ausbildung und in der zweiten Stufe als hormonellen Wirkstoff
lediglich ein Estrogenpräparat enthält, wobei die erste Stufe mindestens 25 und
10 maximal 77 diskrete tägliche oder kontinuierliche Dosierungseinheiten und die zweite
Stufe 5, 6 oder 7 tägliche diskrete oder kontinuierliche Dosierungseinheiten umfaßt
und die Gesamtzahl der Tageseinheiten gleich der Gesamtzahl der Tage des
gewünschten, mindestens 30 und maximal 84 Tage langen, Zyklus ist,
- 15 2. Kombinationspräparat nach Anspruch 1, dadurch gekennzeichnet, daß die erste
Stufe 25 oder 26 tägliche Dosierungseinheiten umfaßt.
3. Kombinationspräparat nach Anspruch 1, dadurch gekennzeichnet, daß die erste
Stufe mindestens 28 und maximal 84 tägliche Dosierungseinheiten umfaßt.
- 20 4. Kombinationspräparat nach Anspruch 3, dadurch gekennzeichnet, daß die erste
Stufe 28 tägliche Dosierungseinheiten umfaßt.
5. Kombinationspräparat nach Anspruch 3, dadurch gekennzeichnet, daß die erste
25 Stufe 28 plus 7 oder 28 plus ein Mehrfaches von 7 tägliche Dosierungseinheiten
umfaßt.
6. Kombinationspräparat nach Anspruch 1, dadurch gekennzeichnet, daß die
zweite Stufe 7 tägliche Dosierungseinheiten umfaßt.
- 30 7. Kombinationspräparat nach Anspruch 2, dadurch gekennzeichnet, daß die
zweite Stufe 5 oder 6 tägliche Dosierungseinheiten umfaßt, so daß das
Kombinationspräparat insgesamt 30 oder 31 tägliche Dosierungseinheiten aufweist.
- 35 8. Kombinationspräparat nach einem der vorstehenden Ansprüche 1 bis 7,
dadurch gekennzeichnet, daß das Estrogen der ersten Stufe aus der Gruppe der
Verbindungen

17 β -Estradiol,

Ethinylestradiol und
17 β -Estradiolvalerat

und das Gestagen aus der Gruppe der Verbindungen

- 5 Dienogest
 Gestoden,
 Levonorgestrel,
 Desogestrel,
 3-Ketodesogestrel,
10 \ Dröspironenon, ~
 Cytoeronacetat,
 Norgestimat und
 Norethisteron sowie

15 das Estrogen der zweiten Hormonkomponente aus der Gruppe der Verbindungen.

17 β -Estradiol,
Ethinylestradiol und
17 β -Estradiolvalerat

20 ausgewählt ist.

9. Kombinationspräparat nach einem der vorstehenden Ansprüche 1 bis 8,
dadurch gekennzeichnet, daß das Estrogen der ersten Stufe in jeder täglichen
25 Dosierungseinheit in einer Dosis von

1.0 bis 6.0 mg 17 β -Estradiol,
— 0.015 bis 0.025 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

30 und das Gestagen in jeder täglichen Dosierungseinheit in einer Dosis von

- 1.0 bis 3.0 mg Dienogest
0.05 bis 0.075 mg Gestoden
0.05 bis 0.125 mg Levonorgestrel
35 0.06 bis 0.15 mg Desogestrel
0.06 bis 0.15 mg 3-Ketodesogestrel
— 1.0 bis 3.0 mg Drospironenon
1.0 bis 2.0 mg Cytoeronacetat
0.2 mg bis 0.3 mg Norgestimat

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0.35 bis 0.75 mg Norethisteron

enthalten ist.

- 5 10. Kombinationspräparat nach einem der vorstehenden Ansprüche 1 bis 9, dadurch gekennzeichnet, daß in der zweiten Stufe in jeder täglichen Dosierungseinheit eine Menge von

10 1.0 bis 6.0 mg 17 β -Estradiol,
0.002 bis 0.04 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

enthalten ist.

- 15 11. Kontrazeptionelles Kit enthaltend mindestens 30 tägliche, jeweils mindestens einen hormonellen Wirkstoff enthaltende, Dosierungseinheiten mit einer ersten und einer zweiten Stufe,
welches in seiner ersten Stufe als hormonellen Wirkstoff in Kombination ein Estrogen- und in mindestens zur Ovulationshemmung ausreichender Dosierung ein
20 Gestagenpräparat in einstufiger Ausbildung und in der zweiten Stufe als hormonellen Wirkstoff lediglich ein Estrogenpräparat enthält, wobei die erste Stufe mindestens 25 und maximal 77 diskrete tägliche oder kontinuierliche Dosierungseinheiten und die zweite Stufe 5, 6 oder 7 tägliche diskrete oder kontinuierliche Dosierungseinheiten umfaßt und die Gesamtzahl der Tageseinheiten gleich der Gesamtzahl der Tage des
25 gewünschten, mindestens 30 und maximal 84 Tage langen, Zyklus ist.

12. Kontrazeptionelles Kit nach Anspruch 11, dadurch gekennzeichnet, daß dessen erste Stufe 25 oder 26 tägliche Dosierungseinheiten umfaßt.

- 30 13. Kontrazeptionelles Kit nach Anspruch 11, dadurch gekennzeichnet, daß die erste Stufe mindestens 28 und maximal 84 tägliche Dosierungseinheiten umfaßt.

14. Kontrazeptionelles Kit nach Anspruch 13, dadurch gekennzeichnet, daß die erste Stufe 28 tägliche Dosierungseinheiten umfaßt.

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15. Kontrazeptionelles Kit nach Anspruch 13, dadurch gekennzeichnet, daß die erste Stufe 28 plus 7 oder 28 plus ein Mehrfaches von 7 tägliche Dosierungseinheiten umfaßt.

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16. Kontrazeptionelles Kit nach Anspruch 11, dadurch gekennzeichnet, daß die zweite Stufe 7 tägliche Dosierungseinheiten umfaßt.
17. Kontrazeptionelles Kit nach Anspruch 12, dadurch gekennzeichnet, daß die
5 zweite Stufe 5 oder 6 tägliche Dosierungseinheiten umfaßt, so daß das Kit insgesamt 30 oder 31 tägliche Dosierungseinheiten aufweist.
18. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 11 bis 17, dadurch gekennzeichnet, daß das Estrogen der ersten Stufe aus der Gruppe der
10 Verbindungen

17 β -Estradiol,
Ethinylestradiol und
17 β -Estradiolvalerat

15

und das Gestagen aus der Gruppe der Verbindungen

Dienogest
Gestoden,
Levonorgestrel,
20 Desogestrel,
3-Ketodesogestrel,
Drospironenon,
Cyptoeronacetat,
Norgestimat und
25 Norethisteron sowie

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25

das Estrogen der zweiten Hormonkomponente aus der Gruppe der Verbindungen.

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17 β -Estradiol,
Ethinylestradiol und
17 β -Estradiolvalerat

ausgewählt ist.

- 35 19. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 11 bis 18, dadurch gekennzeichnet, daß das Estrogen der ersten Stufe in jeder täglichen Dosierungseinheit in einer Dosis von

1.0 bis 6.0 mg 17 β -Estradiol,

→ 0.015 bis 0.025 mg Ethinylestradiol,
1.0 bis 4.0 mg 17β-Estradiolvalerat

und das Gestagen in jeder täglichen Dosierungseinheit in einer Dosis von

- 5 1.0 bis 3.0 mg Dienogest
 0.05 bis 0.075 mg Gestoden
 0.05 bis 0.125 mg Levonorgestrel
 0.06 bis 0.15 mg Desogestrel
 0.06 bis 0.15 mg 3-Ketodesogestrel
10 — 1.0 bis 3.0 mg Drospironenon
 1.0 bis 2.0 mg Cytoeronacetat
 0.2 mg bis 0.3 mg Norgestimat
 0.35 bis 0.75 mg Norethisteron

15 enthalten ist.

20. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 11 bis 19, dadurch gekennzeichnet, daß in der zweiten Stufe in jeder täglichen Dosierungseinheit eine Menge von

- 20 1.0 bis 6.0 mg 17β-Estradiol,
 — 0.002 bis 0.04 mg Ethinylestradiol,
 1.0 bis 4.0 mg 17β-Estradiolvalerat

25 enthalten ist.

21. Kontrazeptionelles Kit enthaltend mindestens 30 tägliche, jeweils mindestens einen hormonellen Wirkstoff enthaltende, Dosierungseinheiten mit einer ersten und einer zweiten Stufe,

- 30 welches in seiner ersten Stufe als hormonellen Wirkstoff in Kombination ein Estrogen- und in mindestens zur Ovulationshemmung ausreichender Dosierung ein Gestagenpräparat in einstufiger Ausbildung und in seiner zweiten Stufe als hormonellen Wirkstoff lediglich ein Estrogenpräparat enthält, wobei die erste Stufe mindestens 25 und maximal 77 diskrete tägliche oder kontinuierliche
35 Dosierungseinheiten und die zweite Stufe 5, 6 oder 7 tägliche diskrete oder kontinuierliche Dosierungseinheiten umfaßt und die Gesamtzahl der Tageseinheiten gleich der Gesamtzahl der Tage des gewünschten, mindestens 30 und maximal 84 Tage langen, Zyklus ist,

wobei sich die Dosierungseinheiten der ersten Stufe teilweise in sich periodisch wiederholenden Untereinheiten befinden, die räumlich und/oder durch andere Markierungen voneinander getrennt sind.

5 22. Kontrazeptionelles Kit nach Anspruch 21, dadurch gekennzeichnet, daß sich die Dosierungseinheiten frühestens ab der 26. täglichen Dosierungseinheit in Untereinheiten befinden.

23. Kontrazeptionelles Kit nach Anspruch 21 oder 22, dadurch gekennzeichnet,
10 daß sich die einzelnen Untereinheiten durch Perforierungen oder andere zum Abtrennen geeignete Vorrichtungen voneinander trennen lassen.

24. Kontrazeptionelles Kit nach einem der Ansprüche 21, 22 oder 23, dadurch gekennzeichnet, daß die separaten Untereinheiten jeweils 7 Dosierungseinheiten
15 enthalten.

25. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 21 bis 24, dadurch gekennzeichnet, daß die erste Stufe 28 plus 7 oder 28 plus ein Mehrfaches von 7 tägliche Dosierungseinheiten umfaßt.

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26. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 21 bis 25, dadurch gekennzeichnet, daß die zweite Stufe 7 tägliche Dosierungseinheiten umfaßt.

27. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 21 bis 26,
25 dadurch gekennzeichnet, daß das Estrogen der ersten Stufe aus der Gruppe der Verbindungen

17 β -Estradiol,
Ethinylestradiol und
30 17 β -Estradiolvalerat

und das Gestagen aus der Gruppe der Verbindungen

Dienogest
Gestoden,
35 Levonorgestrel,
Desogestrel,
3-Ketodesogestrel,
Drospironenon,
Cyptoeronacetat,

Norgestimat und
Norethisteron sowie

das Estrogen der zweiten Hormonkomponente aus der Gruppe der Verbindungen.

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17 β -Estradiol,
Ethinylestradiol und
17 β -Estradiolvalerat

10 ausgewählt ist.

28. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 21 bis 27,
dadurch gekennzeichnet, daß das Estrogen der ersten Stufe in jeder täglichen
Dosierungseinheit in einer Dosis von

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1.0 bis 6.0 mg 17 β -Estradiol,
0.015 bis 0.025 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

20 und das Gestagen in jeder täglichen Dosierungseinheit in einer Dosis von

1.0 bis 3.0 mg Dienogest
0.05 bis 0.075 mg Gestoden
0.05 bis 0.125 mg Levonorgestrel
0.06 bis 0.15 mg Desogestrel
0.06 bis 0.15 mg 3-Ketodesogestrel
1.0 bis 3.0 mg Drospironenon
1.0 bis 2.0 mg Cyproteronacetat
0.2 mg bis 0.3 mg Norgestimat
0.35 bis 0.75 mg Norethisteron

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enthalten ist.

29. Kontrazeptionelles Kit nach einem der vorstehenden Ansprüche 21 bis 28,
dadurch gekennzeichnet, daß in der zweiten Stufe in jeder täglichen
35 Dosierungseinheit eine Menge von

1.0 bis 6.0 mg 17 β -Estradiol,
0.002 bis 0.04 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

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enthalten ist.

30. Methode zur Kontrazeption bei weiblichen Säugetieren umfassend eine
5 mindestens 30 und maximal 84tägige sequentielle Gabe täglicher Dosierungseinheiten
von mindestens 25 und maximal 77 diskreter täglicher oder kontinuierlicher
Dosierungseinheiten während einer ersten Stufe und 5, 6 oder 7 täglicher diskreter
oder kontinuierlicher Dosierungseinheiten während einer zweiten Stufe enthaltend in
der ersten Stufe als hormonellen Wirkstoff in Kombination ein Estrogen- und in
10 mindestens zur Ovulationshemmung ausreichender Dosierung ein Gestagenpräparat
in einstufiger Ausbildung und in der zweiten Stufe als hormonellen Wirkstoff
lediglich ein Estrogenpräparat.
31. Methode nach Anspruch 30, dadurch gekennzeichnet, daß in der ersten Stufe
15 25 oder 26 tägliche Dosierungseinheiten gegeben werden.
32. Methode nach Anspruch 30, dadurch gekennzeichnet, daß in der ersten Stufe
mindestens 28 und maximal 77 tägliche Dosierungseinheiten gegeben werden.
- 20 33. Methode nach Anspruch 32, dadurch gekennzeichnet, daß in der ersten Stufe
28 tägliche Dosierungseinheiten gegeben werden.
34. Methode nach Anspruch 32, dadurch gekennzeichnet, daß in der ersten Stufe
28 plus 7 oder 28 plus ein Mehrfaches von 7 tägliche Dosierungseinheiten gegeben
25 werden.
35. Methode nach einem der vorstehenden Ansprüche 30 bis 34, dadurch
gekennzeichnet, daß in der zweiten Stufe 7 tägliche Dosierungseinheiten gegeben
werden.
- 30 36. Methode nach Anspruch 31, dadurch gekennzeichnet, daß in der zweiten Stufe
5 oder 6 tägliche Dosierungseinheiten gegeben werden.
37. Methode nach einem der vorstehenden Ansprüche 30 bis 36, dadurch
35 gekennzeichnet, daß das Estrogen in den Dosierungseinheiten der ersten Stufe aus der
Gruppe der Verbindungen

17 β -Estradiol,
Ethinylestradiol und

17 β -Estradiolvalerat

und das Gestagen aus der Gruppe der Verbindungen

- 5 Dienogest
 Gestoden,
 Levonorgestrel,
 Desogestrel,
 3-Ketodesogestrel,
 Drospironenon,
10 Cytoeronacetat,
 Norgestimat und
 Norethisteron sowie

das Estrogen in den Dosierungseinheiten der zweiten Hormonkomponente aus der
15 Gruppe der Verbindungen.

17 β -Estradiol,
Ethinylestradiol und
17 β -Estradiolvalerat

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ausgewählt ist.

38. Methode nach einem der vorstehenden Ansprüche 30 bis 37, dadurch
gekennzeichnet, daß das Estrogen der ersten Stufe in jeder täglichen
25 Dosierungseinheit in einer Dosis von

1.0 bis 6.0 mg 17 β -Estradiol,
0.015 bis 0.025 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

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und das Gestagen in jeder täglichen Dosierungseinheit in einer Dosis von

- 1.0 bis 3.0 mg Dienogest
0.05 bis 0.075 mg Gestoden
0.05 bis 0.125 mg Levonorgestrel
35 0.06 bis 0.15 mg Desogestrel
 0.06 bis 0.15 mg 3-Ketodesogestrel
 1.0 bis 3.0 mg Drospironenon
 1.0 bis 2.0 mg Cytoeronacetat
 0.2 mg bis 0.3 mg Norgestimat

0.35 bis 0.75 mg Norethisteron

gegeben wird.

- 5 39. Methode nach einem der vorstehenden Ansprüche 30 bis 38, dadurch gekennzeichnet, daß in der zweiten Stufe in jeder täglichen Dosierungseinheit eine Menge von

10 1.0 bis 6.0 mg 17 β -Estradiol,
0.002 bis 0.04 mg Ethinylestradiol,
1.0 bis 4.0 mg 17 β -Estradiolvalerat

gegeben wird.

- 15 40. Methode nach einem der Ansprüche 30 bis 39 worin das weibliche Säugetier ein homo sapiens sapiens ist.

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(21) International Application Number: PCT/EP96/04261 (22) International Filing Date: 30 September 1996 (30.09.96) (30) Priority Data: 08/535,402 28 September 1995 (28.09.95) US (71) Applicant: SCHERING AKTIENGESELLSCHAFT [DE/DE]; Müllerstrasse 178, D-13353 Berlin (DE). (72) Inventors: ELLIESEN, Jörg; Elsestrasse 12, D-13467 Berlin (DE). RIEDL, Jutta; Flensburger Strasse 14, D-10557 Berlin (DE).	(81) Designated States: AL, AU, BB, BG, BR, BY, CA, CN, CZ, EE, GE, HU, IL, IS, JP, KE, KR, LK, LR, LT, LV, MG, MK, MN, MX, NO, NZ, PL, RO, RU, SG, SI, SK, TR, TT, UA, UG, UZ, VN, ARIPO patent (KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>Without international search report and to be republished upon receipt of that report.</i>	
(54) Title: HORMONE REPLACEMENT THERAPY METHOD AND HORMONE DISPENSER		
(57) Abstract Varying the daily dose of either or both of the estrogen and the progestogen administered for hormone replacement therapy (HRT) is readily and inexpensively accomplished, without the necessity of the physician prescribing a new product each time the daily dose of the estrogen or progestogen is changed, by administering preferably transdermally the estrogen and progestogen contained in separate extrudable pharmaceutical compositions from a dispenser which contains means, preferably adjustable only by the attending physician or dispensing pharmacist, for varying the volume of either or both of the respective compositions which is dispensed as a single dose from the dispenser in response to a defined digital dispensing manipulation of the dispenser, thereby facilitating optimal compliance to a combination HRT with individually adjusted dosages of the estrogen and progestogen.		

WHAT IS CLAIMED IS:

1. An HRT method of self-administration by a human female of successive doses of an estrogen and a plurality of successive doses of a progestogen, wherein the female dispenses and self-administers successive individual doses of the estrogen and of the progestogen over a protracted period of time, during which period of time the amount of at least one of which doses is changed, which method comprises the steps of:

a) dispensing to a delivery surface, from a first source of a first semisolid of viscous liquid pharmaceutically acceptable composition containing a plurality of non-discrete doses of the estrogen, a metered volume of the first composition as a single dose of the estrogen;

b) concurrently dispensing to the delivery surface, from a second source of a second semisolid or viscous liquid pharmaceutically acceptable composition containing a plurality of non-discrete doses of the estrogen, a metered volume of the second composition as a single dose of the progestogen;

c) transporting the dispensed doses of the first and second compositions from the delivery surface simultaneously to the site of administration of the estrogen and progestogen and administering the dispensed estrogen and progestogen thereat;

d) repeating steps a) - c) on a predetermined schedule; and

e) periodically, in response to the physiological symptoms associated with menopause or the HRT currently exhibited by the female, changing the metered volume of at least one of the first and second compositions which is dispensed onto the delivery surface as a single dose thereof.

2. A method according to claim 1, wherein the predetermined schedule of Step d) is daily.

3. A method according to claim 1, wherein the site of administration is an area of the skin of the female and the first and second pharmaceutically acceptable compositions are adapted to transdermal administration.

4. A method according to claim 3, wherein the delivery surface is the female's hand and the compositions thereon are administered at the selected site of transdermal administration on the skin of the female by rubbing the hand against the the skin.

5. A method according to claim 1, wherein the pharmaceutical compositions are semi-solids which are dispensed as extrudates.

6. A method according to claim 1, wherein the first and second pharmaceutically acceptable compositions are dispensed simultaneously from the respective sources thereof.

7. A method according to claim 1, wherein the dispensed doses of the first and second pharmaceutically acceptable compositions are mixed before being dispensed onto the delivery surface.

8. A method according to claim 1, wherein the predetermined schedule of Step d) is daily; wherein the site of administration is the skin of the female and the first and second pharmaceutically acceptable compositions are adapted to transdermal administration; wherein the compositions thereon are delivered to the selected site of transdermal administration on the skin of the female by rubbing; wherein the pharmaceutical compositions are semi-solids and are dispensed as extrudates simultaneously from the respective sources thereof; and wherein the metered doses of the first and second pharmaceutically acceptable compositions are mixed before being extruded onto the delivery surface.

9. A method according to claim 1, wherein the source of the first pharmaceutical composition is a first compartment of a unitary dispensing means and the source of the second pharmaceutical composition is a second compartment of the dispensing means; wherein the first and second pharmaceutical compositions are first extruded from their respective compartments into first and second metering chambers, respectively; and wherein the contents of the metering chambers are then formed into a single extrudate which is then dispensed from the dispensing means.

10. A method according to claim 1, wherein either the length of the extrudates of the first and second pharmaceutical compositions which are extruded from the respective sources thereof is constant and the dosage of the hormones therein is changed in Step (e) by changing the diameter of one or both of the extrudates, or wherein the diameter of the extrudates of the first and second pharmaceutical compositions which are extruded from the respective sources thereof is constant and the dosage of the hormones therein is changed in Step (e) by changing the length of one or both of the extrudates.

11. A method according to claim 1, wherein the estrogen is ethinyl estradiol or estradiol or an ester thereof, estrone, estrone sulfate or conjugated estrogens and the progestogen is micronized progesterone, norethindrone or esters thereof, norgestrel, chlormadinone acetate, cyproterone acetate, desogestrel, 3-ketodesogestrel, drospirenone, norgestimate, levo-norgestrel or gestodene.

12. A method according to claim 1, wherein the estrogen is ethinyl estradiol or 17 β -estradiol and the progestogen is gestodene, levonorgestrel or 3-ketodesogestrel.

13. A method according to claim 12, wherein the predetermined schedule of Step d) is daily; wherein the pharmaceutical compositions are semi-solids; wherein the pharmaceutical compositions are metered from a dispenser in which the first and second containers both are housed, which the dispenser comprises mechanical means for changing the volumes of the pharmaceutical compositions which are dispensed from the containers in response to a defined constant digital manipulation employed to extrude from the container.

14. A dispenser adapted for dispensing and self-administering a plurality of successive individual doses of an estrogen and a progestogen from a dispenser containing multiple doses of thereof in separate extrudable pharmaceutically acceptable compositions, in metered single dosage amounts which can be varied by mechanically modifying the dispenser, which comprises:

a) a first container comprising:

(i) a first chamber containing a plurality of doses of the estrogen in a first extrudable pharmaceutically acceptable composition;

(ii) a discharge port through which a volume of the first pharmaceutical composition in the first chamber can be discharged therefrom when discharging pressure is applied to the first container;

(iii) means for applying discharging pressure to the first chamber;

b) a second container comprising:

(i) a second chamber containing a plurality of doses of the progestogen in a second extrudable liquid pharmaceutically acceptable composition;

(ii) a discharge port through which a volume of the second extrudable pharmaceutical composition in the second chamber can be discharged therefrom when discharging pressure is applied to the second container;

(iii) means for applying discharging pressure to the second chamber;

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c) a first metering means adapted to receive a volume of the first extrudable pharmaceutical composition extruded thereinto from the first chamber and digitally reciprocally convertible from a receiving mode, where it communicates with the first chamber through the first port and where it can be filled with a volume of the first extrudable pharmaceutical composition, to a discharging mode, where a volume of the first extrudable pharmaceutical composition therein can be discharged therefrom and dispensed from the dispenser, which first metering means comprises:

(i) a first adjusting means for varying the volume of the first extrudable pharmaceutical composition which can be extruded thereinto;

d) a second metering means adapted to receive a volume of the second extrudable pharmaceutical composition extruded thereinto from the second chamber and digitally reciprocally convertible from a receiving mode, where it communicates with the second chamber through the second port and where it can be filled with a volume of the second extrudable pharmaceutical composition, to a discharging mode, where a volume of the second extrudable pharmaceutical composition therein can be discharged therefrom and dispensed from the dispenser, which second metering means comprises:

(i) a second adjusting means for varying the volume of the second extrudable pharmaceutical composition which can be extruded thereinto as a single dose of the progestogen; and

e) dispensing means for dispensing a volume of the first pharmaceutical composition and a volume of the second pharmaceutical composition from the first and second metering means, respectively, from the dispenser as a single dose of the estrogen and the progestogen.

15. A dispenser according to claim 14 which comprises means for blending the single dose of the estrogen and the single dose of the progestogen discharged from the first and second metering means into a single extrudate prior to being dispensed from the dispenser.

16. A dispenser according to claim 14, wherein the first and second adjusting means are positioned internally in the dispenser and not readily accessible by an individual dispensing single doses of the estrogen and the progestogen therefrom.

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17. A dispenser according to claim 14, wherein the estrogen is ethinyl estradiol or estradiol or an ester thereof, estrone, estrone sulfate or conjugated estrogens and the progestogen is micronized progesterone, norethindrone or esters thereof, norgestrel, chlormadinone acetate, cyproterone acetate, desogestrel, 3-ketodesogestrel, drospirenone, norgestimate, levo-norgestrel or gestodene.

18. A dispenser according to claim 14, wherein the estrogen is ethinyl estradiol or 17 β -estradiol and the progestogen is gestodene, levonorgestrel or 3-ketodesogestrel.

19. A dispenser according to claim 18, which comprises means for blending the single dose of the estrogen and the single dose of the progestogen discharged from the first and second metering means into a single extrudate prior to being dispensed from the dispenser, wherein the estrogen is ethinyl estradiol or 17 β -estradiol, the progestogen is gestodene, levonorgestrel or 3-ketodesogestrel.

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Numbering	Prior art document
D1	WO 97/11680
D2	US 5,922,349
D3	DE 3 022 337 C2
D4	WO 98/04269
D5	Fotherby, <i>Contraception</i> , 1996;54;59-69
D6	EP 0 461 290 A1
D7	Chaumell, <i>Meth Find Expt Clin Pharmacol</i> , 1998;20;211-215
D8	McInnes et al, <i>J Clin Pharmacol</i> , 1982;22;410-417
D9	Fuhrmann et al, <i>Contraception</i> , 1996;54;243-251
D10	US 4,196,188
D11	Maxson et al, <i>Fertility and Sterility</i> , 1985;44;622-626
D12	US 4,927,816
D13	De Villiers, <i>J Pharm Biomed Anal</i> , 1995;13;191-198
D14	US 4,826,831
D15	WO 95/17195
D16	WO 97/01342
D17	US 08/535,402 (earlier application of D1 and D2)
D18	Nikisch et al, <i>Tetrahedron Letters</i> , 1986;27;5463-5466
D19	Garret et al, <i>J Pharm Sci</i> , 1971;60;1801-1809
D20	Pramar et al, <i>J Pharm Sci</i> , 1991;80;551-553
D21	Kincl et al, <i>J Steroid Biochem</i> , 1978;9;83-84
D22	Hargrove et al, <i>Am J Obstet Gynecol</i> , 1989;948-951
D23	Supplementary experimental data

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NOVELTY

The opponent alleges that independent claims 1 and 16 lack novelty over D1 as well as D2.

More particularly, the opponent states that the text on page 15 of D1 (second paragraph), and the text of D2 (column 10, lines 9-10 and 27) disclose drospirenone in micronised form.

We completely disagree with the opponent's statement.

It is respectfully submitted that the opponent deliberately has chosen to read the above-mentioned paragraphs in isolation of the remaining text of the documents.

As explained in further detail below, D1 as well as D2 merely teach "micronised progesterone" - not "micronised drospirenone", since only the natural gestagen, progesterone, is further characterised by the term "micronised" in D1 and D2. Accordingly, neither D1 nor D2 anticipates claims 1 and 16 of the opposed patent.

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D10

United States Patent [19]

[11] 4,196,188

Besins

[45] Apr. 1, 1980

[54] ORALLY ADMINISTRABLE FORM OF
PROGESTERONE

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[21] Appl. No.: 759,277

[22] Filed: Jan. 12, 1977

[51] Int. Cl.² A61K 9/48

[52] U.S. Cl. 424/37; 424/242;
424/243

[58] Field of Search 424/37, 238, 243

[56] References Cited

U.S. PATENT DOCUMENTS

3,284,303 11/1966 Meli 424/242
3,342,682 9/1967 Broch 424/238

3,828,106 8/1974 Rodez 424/242 X
3,862,311 1/1975 Leeson 424/242 X

Primary Examiner—Shep K. Rose
Attorney, Agent, or Firm—Gifford, Chandler,
VanOphem, Sheridan & Sprinkle

[57] ABSTRACT

The present invention relates to an orally active pharmaceutical preparation comprised of progesterone, finely ground such that 80% of the progesterone particles are about 1-15 microns in size, suspended in an excipient and contained in capsules. The drug effects increased progestational and contraceptive activity at this particle size.

1 Claim, No Drawings

ORALLY ADMINSTRABLE FORM OF
PROGESTERONE

BACKGROUND OF THE INVENTION

1. Field of the Invention

Progesterone is a well-known natural hormone which is, in practice, only administered by injection, and otherwise in the form of delayed release derivatives, in cases of hormone deficiency of the corpus luteum, especially in cases of infertility.

2. Description of the Prior Art

The administration of progesterone by the oral route has been abandoned because of its destruction in the digestive tract which occurs more rapidly than its absorption, to a point such that even daily doses of one or two grams are still insufficient to give rise to a net progestational action.

As a result, up to the present day, the only method for obtaining progestational action by oral administration has consisted in using synthetic derivatives such as medroxyprogesterone, norethisterone, methylestrenolone, chlormadinone acetate, 6-dehydroretroprogesterone or lynestrenol.

However, these derivatives suffer from a number of disadvantages. In the first case, their activities are not wholly identical with those of progesterone. Thus, certain of the derivatives possess a notable androgenic activity, giving rise to a long term risk of virilisation, as well as a risk of masculinisation of the foetus in the case of treatment in the course of pregnancy. Others of the derivatives possess a long term estrogenic activity.

Further, in contradistinction to progesterone, certain of the derivatives do not possess anti-estrogenic activity, anti-aldosteronic activity or anti-ovulatory activity.

Secondly, these derivatives give rise to problems of tolerance, which manifest themselves as digestive troubles, the onset of migraine or headaches and even the risk of venous thrombosis. Accordingly, the Applicant has carried out research with a view to providing a form of progesterone which is effectively administrable by the oral route, whilst giving rise to all its properties, and without secondary effects, as well as a practical method for preparing this form of progesterone and presenting it to the patient, on a commercial scale.

SUMMARY OF THE INVENTION

In accordance with the invention the therapeutically effective form for oral administration of progesterone consists of particles micronised to a particle size of less than 15 microns.

The invention also provides, as indicated above, a composition for the therapeutically effective administration of progesterone, for its usual applications, characterized in that it contains, as active constituent, micronised progesterone of which at least 80% of the particles have a particle size between 1 and 15 microns, together with an appropriate vehicle or excipient which, as will be discussed below, is preferably an oil or an oily product.

DESCRIPTION OF THE PRESENT INVENTION

Thus it has been found that if progesterone is micronised to such a particle size, and administered by the oral route, it is assimilated at a rate greater than the rate of its destruction in the digestive tract, thereby exercising

ing its progestational activity whilst being free from all unwanted secondary side effects.

More particularly, the applicant has carried out experiments to determine the critical character of the particle size in both animal and human trials.

The particle size analysis of the progesterone under trial was carried out using a Coulter Counter, Model TA 2, which is an automatic apparatus for analysing particle size distribution of powders. This apparatus makes it possible to detect particles independently of their form, their density or their colour, and the measure of the size is a direct function of their volume and thus of their mass. The apparatus operates over such a scale that it is possible to differentiate between particles whose mass varies from 0.04 microns to several hundreds of microns. The analysis is carried out on several hundred thousands of particles as a result of which it is possible to carry out a valid check on the particle size distribution.

These various advantages give the results of the analysis obtained by this method an exactitude, a precision and a reproducibility which cannot be obtained by other conventional methods for granulometric analysis.

Experience shows that for the application envisaged, the progesterone particles should have a particle size of less than 15 microns.

The test used to establish this limit consists in measuring the level of progesterone in plasma by the radio-immunological method using double isotopic labelling and chromatography (Scholler-Mauvais-Jarvis).

Animal Trials

In the course of pharmacological trials on animals, there were measured the progesterone plasma levels after administration of different doses of progesterone having different particle sizes by the oral route to castrated rats. The two dosage rates used were 5 mg/kg and 10 mg/kg.

The two types of progesterone had particle sizes of 5 microns and of 100 microns. Administration was effected by forced feeding of fasting animals with the progesterone suspended in carboxymethyl cellulose. Blood samples were taken two hours and four hours after administration of the progesterone. The plasma progesterone levels were measured by the radio-immunological method using an antiserum from the Institut Pasteur. The results obtained are given in Table I below.

TABLE I

		2h	4h
5 mg/Kg	100µ	22.5 ± 1	15 ± 2.5
	5 µ	31 ± 6.5	34 ± 7.5
10 mg/Kg	100µ	28.5 ± 3.5	10 ± 5
	5 µ	32.5 ± 5	24 ± 4

The following comments may be made in view of these results.

1. The plasma concentrations measured are not (at the doses used) a function of the dose administered; the results are practically identical with the two doses used of 5 mg/Kg and 10 mg/KG.

2. At a dose of 5 mg/Kg the plasma concentrations are higher when the progesterone is micronised to 5 microns than when it is micronised to 100 microns. At this dosage and at the time of four hours the difference is significant.

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3. At the two dosages employed, 5 mg/Kg and 10 mg/Kg, the plasma concentrations are comparable after two hours and four hours for the progesterone micronised to 5 microns. On the contrary, for the progesterone micronised to 100 microns, the plasma concentrations are significantly less four hours after administration than at two hours after administration.

II—Human Trials

In man, progesterone micronised in accordance with the invention is administered by the oral route in the form, for example, of gelules or soft gelatine capsules at doses of from 100 to 200 mg to exercise its progestational and contraceptive activity.

The results of these trials, namely the plasma levels with different doses of micronised progesterone, the pharmaceutical presentation being as described above, are given in Table 2 below.

TABLE 2

		2h	6h	24h
100µ	200 mg	16	7	0.6
20µ	200 mg	27	7.4	3.5
	100 mg	14	8	4
10µ	200 mg	78	17	2.5
5µ	200 mg	62	7	1.6
After 3 months storage		17	3.5	3
10µµ	200 mg	25.3	9.2	6.8
	100 mg	8.8	5.4	8.6

It will be clearly seen from Table 2 that the same observations as given for the results summarised in Table 1, apply.

It is convenient to note that the method of preparation of the micronised progesterone compositions in accordance with the invention is of importance.

In the first case, the technique employed for micronisation of the progesterone implies a certain number of important conditions; thus in order to obtain a given, constant granulometry for each operation, it is necessary to start from a particular crystalline form of progesterone, obtained by recrystallization, whatever may be the physical characteristics of the starting progesterone.

To this end, in a first stage, the progesterone is dissolved, at 60° C. in acetone at a ratio of one part of progesterone to two volumes of acetone. The solution is then filtered and the progesterone is recrystallized by progressive cooling with stirring.

The evolution of temperatures during the course of the cooling must be rigorously followed in order to obtain a regular constant crystal form. The final temperature is 5° C. The yield of this crystallization is about 90%. The crystals obtained are dried under vacuum until all the acetone has been removed. In the second step, the micronisation proper should be carried out in a high speed rotor mill containing one part of arachid oil (codex) for one part of progesterone. The time of milling or grinding will be a function of the capacity and type of apparatus used. This must be calibrated with increasing grinding times to obtain a given granulometry. The length of grinding may vary from half an hour to three hours.

The grinder or mill must be provided with a cooling system to maintain the temperature between 25° and 30° C. All parts of the grinder in contact with the progesterone should be made of stainless steel. It is preferable to carry out grinding under nitrogen.

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The progesterone thus micronised must then be formulated in the appropriate pharmaceutical form. In this regard, it is convenient to note that compressed forms such as dragees, tablets, pilules, etc. may not be used since all these dry oral dosage unit forms necessitate at one stage or another in their preparation a more or less great compression of the ingredients. It is known that compression modifies the granulometric distribution of the starting particles. Thus, if one starts with an initial powder having a fixed particle size, disintegration after compression, even light compression, of the particles gives rise to very different particle sizes and hence gives rise to a very different bioavailability.

Accordingly, the micronised progesterone in accordance with the invention should be presented in the form of gelules which overcome the above disadvantages since one may thus reduce or even eliminate all compression of the product during the course of filling of the gelule. However, when micronised to a particle size of less than twenty microns, the product becomes very hygroscopic and electrostatic and is thus very difficult to handle. Further, in the course of time, a modification of the granulometric distribution is noted in that the average particle size increases, leading to a significant reduction in the plasma levels after absorption, as shown in Table 3 below.

TABLE 3

Particle size	DOSE	2h	6h
5µ fresh product	200 mg	62	7
after 3 months storage		17	3.5

All these problems can be overcome, however, if the quantity of micronised progesterone to be administered only represents a small percentage of the total weight of the gelule. However, with regard to the dose of the order of from 100 to 200 mg necessary to obtain plasma levels corresponding to the desired therapeutic activity, the major part of the contents of the gelule must comprise the active principal.

Accordingly, the applicants have been led to another pharmaceutical form which permits the administration, by the oral route, of a liquid or semi-liquid or pasty product, in the absence of water—namely the soft capsule.

In view of the method of production of such soft capsules, the excipient is an oil or other analogous oily product, which permits filling of the capsule by flowing the composition into the capsule. It is for this reason that the process in accordance with the invention, consisting of micronising the progesterone directly in the oily phase, is carried out since this makes it possible to immediately fill the desired product into capsules without the risk of modifying the granulometric distribution of the micronised material in the course of various handling operations and also facilitates storing of the product. Further, micronisation itself is carried out more easily in the oily phase than in the dry or aqueous phase and thus overheating, always undesirable, of the active principle is avoided and the necessity for cooling and lubricating the course of the micronisation is also avoided.

It is convenient to note that the German chemical firm Schering have suggested, among their products, a micronised progesterone but as chemical product, sold as such, without any definite pharmaceutical form,

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without specifying any particular use and, particularly, without indicating any particular properties or applications. Accordingly, this is the first time progesterone has been presented as such as a medicament which may be effectively administered by the oral route in a form capable of exercising all the effects of endogenous progesterone, without secondary effects. On the one hand there is the anticonceptual effect of progestational agents and on the other hand a true substitutive activity in all cases of luteal insufficiency.

The indications for the compositions of the invention are, therefore, the following:

Luteal insufficiency

as evidenced by premenstrual anxiety
dysmenorrhea
functional meno-metrorragia
amenorrhea

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benign mastodynia of mastopathia
Seborrhea, acne, seborrhic alopecia

Endometriosis
Threatened Abortion
Contraception

Having described my invention, many modifications thereto will become apparent to those skilled in the art to which it pertains without departing from the spirit of the invention as defined by the scope of the appended claims.

I claim:

1. An effective orally administrable soft gelatin capsule essentially containing micronised particles of recrystallized progesterone at least 80% having a particle size of from 5 to less than about 15 microns, in an oil vehicle, said micronized particles having been micronized in said oil phase.

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Bioavailability of oral micronized progesterone*

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Progesterone (P) has not been administered orally because of reportedly poor bioavailability and a rapid clearance rate. Unfortunately, the synthetic derivatives, although orally active, have a number of disadvantages and fail to mimic natural P completely. To investigate the bioavailability and short-term toxicity of oral micronized P, a standardized dose of 200 mg of micronized P was administered to nine healthy postmenopausal women and one male subject. Serial determinations of serum P concentrations demonstrated rapid absorption of P. Peak concentrations of P rose from a negligible baseline level to 17.0 ± 4.9 ng/ml at an average of 2.8 ± 0.35 hours after administration. The peak concentrations of P were equivalent to those observed in the midluteal phase in normal control cycles (14.1 ± 2.7 ng/ml). All subjects exhibited significant elevation of P over baseline levels that persisted for at least 6 hours after the single oral dose and returned to initial levels by 24 hours. There was no significant change in estradiol, follicle-stimulating hormone, luteinizing hormone, cortisol, aldosterone, lipids, or hepatic enzymes during the 24-hour study interval. Fertil Steril 44:622, 1985

Progestins are administered for a wide variety of clinical indications, which include oral contraception, adjunctive therapy in menopause, endometrial hyperplasia or carcinoma, endometriosis, dysfunctional uterine bleeding, amenorrhea, luteal phase inadequacy, and the premenstrual syndromes.¹ Unfortunately, the oral route of administration of naturally occurring progesterone (P) has not been practical because of the rapid hepatic metabolism and the poor bioavailability of this steroid.² Thus, synthetic derivatives (e.g., norethindrone acetate, norgestrel, medroxyprogesterone acetate, lynestrenol, ethynodiol diace-

tate, and norethynodrel) have been the only orally active progestational agents available. The synthetic progestins suffer from a number of disadvantages. None of these agents have effects identical to natural P. Certain derivatives exhibit prominent side effects, which include masculinization of the patient or fetus, depression, fluid retention, headaches, alterations in serum lipid profiles, and possible teratogenicity.¹⁻³ In addition, these synthetic progestins demonstrate variable antiestrogenic, estrogenic, and antiovarian activity.¹⁻³

Because of these problems, investigation has been rekindled in the search for an orally active form of natural P. Recent preliminary studies^{1, 4-7} suggest that a micronized form of natural P is readily absorbed orally and may offer a novel alternative to the synthetic agents.

This study was designed for evaluation of the bioavailability and short-term toxicity of oral micronized P in an easily prepared dosage.

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Table 1. Patient Profile^a

Subject no.	Sex	Age	Height	Weight	Age at menopause	Exogenous estrogen	Exogenous progestogen
		yr	in	lbs	yr		
1	F	46	63	139	41	Conjugated estrogen 1.25 mg/day	None
2	F	41	63	128	37	E ₂ valerate in- jections and diethyl- stilbestrol 0.25 mg/ day	None
3	F	54	62.5	152	53	Conjugated estrogen 1.25 mg/day	Medroxyprogesterone acetate 10 mg daily 10 days/month
4	F	61	65	203	33	None	None
5	F	36	64	228	28	None	None
6	F	47	69	136	46	None	None
7	F	38	60	112	26	None	None
8	F	41	65.5	146	37	Conjugated estrogen 1.25 mg/day	None
9	F	37	63	184	29	Conjugated estrogen 1.25 mg/day	None
10	M	47	69	215	—	None	None

^aM, male; F, female.

MATERIALS AND METHODS

Nine healthy postmenopausal women and one male subject volunteered for the study (Table 1). Five women were receiving exogenous estrogens. One woman had previously received progestational agents, but none of the volunteers had taken any progestin within 3 weeks before the onset of the study. The participants were fasting at the onset of the study but were allowed to eat 4 hours after the administration of P. The patients were ambulatory but were without strenuous exercise for the first 6 hours of the study. During the subsequent 18 hours, normal activity and diet were permitted.

P, USP micronized, lot #076UJ, was obtained from The Upjohn Company, Kalamazoo, MI. The particle geometric mean diameter was $11 \pm 1.7 \mu$ (particle size: 90% $< 20 \mu$, 95% $< 10 \mu$, and 71% $< 5 \mu$). Micronized P, 100 mg, was placed into #3 plain gelatin capsules (Delk Pharmacy, Columbia, TN).

With subjects fasting, a 21-gauge heparin lock needle (Abbott Intermittent Infusion Set, #4721, Abbott Laboratories, North Chicago, IL) was inserted intravenously at 9:00 A.M. All subjects received 200 mg of micronized P by mouth at 9:05 A.M. Blood was drawn before P administration and at 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 5.0, 6.0, and 24.0 hours thereafter. Serum was separated by centrifugation, labeled, and frozen for determinations of P, estradiol (E₂), cortisol, dehydroepian-

drosterone sulfate (DHEA-S), follicle-stimulating hormone (FSH), luteinizing hormone (LH), high-density lipoproteins (HDL), triglycerides, cholesterol, serum glutamic pyruvic transaminase (SGPT), bilirubin, and alkaline phosphatase. The blood pressure, pulse, and temperature were monitored at each blood drawing.

Serum alkaline phosphatase, SGPT, bilirubin, HDL, cholesterol, and triglycerides were determined with commercial kits from Mallinckrodt, Inc., St. Louis, MO with the use of the Mallinckrodt Serometer Model 375.

The assays for P, cortisol, FSH, and LH were performed by radioimmunoassay (RIA) with the use of commercial kits obtained from Diagnostic Products Corporation, Los Angeles, CA. Intraassay variations for these hormones were 7.5%, 4.1%, 3.1%, and 3.2%, respectively. The interassay variations were 4.3%, 8.8%, 4.6%, and 5.4%, respectively. The RIAs for E₂ and DHEA-S were performed with kits obtained from Pantex, Santa Monica, CA. Intraassay variations were 5.6% and 6.2%, respectively. Interassay variations were 13.8% and 7.6%, respectively.

Normal midluteal P concentrations for the Diagnostic Products' RIAs were determined in seven healthy women with regular ovulatory cycles. Because luteal phase P concentrations may be pulsatile and vary according to the day of the cycle,⁸ serum from each subject was obtained in the morning on 3 separate days in the midluteal phase. These samples were pooled for each individual patient.

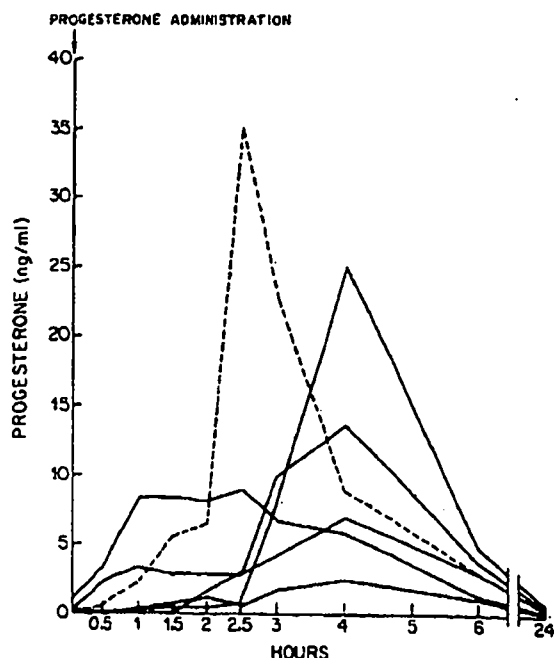


Figure 1
Serial P concentrations after the oral administration of micronized P, 200 mg at time 0; male (---) and female (—) subjects with E_2 concentrations < 50 pg/ml.

Data were analyzed by Student's *t*-test and correlation analysis.

RESULTS

Baseline P concentrations were < 1 ng/ml. In all patients, P levels increased to 2 ng/ml after P administration, reaching a peak of 17.0 ± 4.9 ng/ml (mean $\pm$ standard error of the mean [SEM]) at an average of 2.8 ± 0.35 hours (Figs. 1 and 2 and Table 2). Peak concentrations of P in this group were not significantly different from mean midluteal levels in the control cycles (14.1 ± 2.7 ng/ml).

The rate of absorption was variable (Figs. 1 and 2). P concentrations rose above 1 ng/ml in three patients by 0.5 hours; six patients at 1.0 hours, seven patients at 1.5 hours, and nine patients at 2.0 hours.

All subjects exhibited a significant elevation of P over baseline levels that persisted for at least 6 hours after a single oral dose and that subsequently returned to initial levels by 24 hours (Fig. 3).

Peak P concentrations were positively correlated with serum E_2 concentrations measured con-

comitantly ($r = 0.636$, $P < 0.02$). However, there was no significant change in mean E_2 levels at any of the sampling intervals (Table 2).

No significant change in blood pressure, temperature, pulse rate, FSH, LH, bilirubin, alkaline phosphatase, SGPT, or lipids was observed between the time of baseline determination, peak P concentration, and 24 hours after administration (Table 2).

No major complications were reported in these subjects during the 24-hour study interval. Minor side effects included drowsiness in three women. This symptom was noted between 1 and 3 hours after oral administration of P, within 1 hour of the measurement of peak P concentration in each individual. One subject complained of exacerbated hot flashes at 24 hours. No gastrointestinal side effects during the 24-hour study interval were reported.

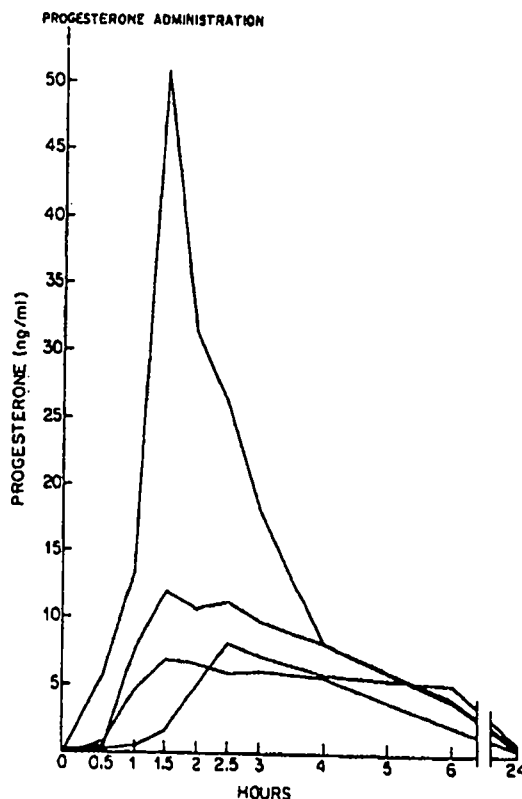


Figure 2
Serial P concentrations after the oral administration of micronized P, 200 mg at time 0. Female subjects with E_2 concentrations > 50 pg/ml.

Table 2. Serum Concentrations of Steroids, Enzymes, and Lipids in Female Subjects ($n = 9$) Receiving Oral Micronized P 200 mg at Time 0

Test	Units ^a	Time				
		0	2	4	6	24
P	ng/ml	0.2 ± 0.1^b	7.5 ± 3.1^c	9.1 ± 2.2^d	3.2 ± 4.1^d	0.6 ± 0.1
E ₂	pg/ml	72 ± 24	57 ± 18	60 ± 18	62 ± 19	56 ± 14
DHEA-S	ng/ml	1603 ± 614	1488 ± 578	1646 ± 611	1713 ± 590	1860 ± 688
FSH	mIU/ml	70 ± 11	63 ± 13	65 ± 11	73 ± 12	73 ± 12
LH	mIU/ml	68 ± 13	69 ± 12	69 ± 12	73 ± 10	74 ± 13
Cortisol	μg/dl	12 ± 3				11 ± 2
Aldosterone	ng/dl	18 ± 4				18 ± 2
Cholesterol ^e	mg/dl	284 ± 19				279 ± 20
HDL ^e	mg/dl	43 ± 4				43 ± 4
Triglycerides ^e	mg/dl	190 ± 23				188 ± 19
Alkaline phosphatase ^e	Bodansky U/dl	4.3 ± 0.8				4.3 ± 0.5
SGPT ^e	IU/l	11 ± 0.2				11 ± 1
Bilirubin ^e	mg/dl	0.2 ± 0.1				0.2 ± 0.1

^aAll values are recorded as mean $\pm$ SEM.

^bStatistical comparison of baseline values to subsequent measurements by Student's *t*-test. All comparisons not significant ($P > 0.05$), except as noted.

^c $P < 0.05$.

^d $P < 0.001$.

^eValues include one male subject ($n = 10$).

DISCUSSION

Natural P has not been used clinically as an oral preparation because of reportedly poor gastrointestinal absorption and a short biologic half-life.^{6,7} However, in 1978, Kincl et al.⁶ demonstrated that nonmicronized P on a lactose carrier in gelatin capsules rapidly increased serum P concentrations in male volunteers. An oral dose of 100 mg of nonmicronized P produced mean serum concentrations that peaked at 2 hours, with a mean of 8.6 ng/ml (range, 3.8 to 13.4 ng/ml), and declined rapidly to 3.2 ng/ml by 4 hours.⁶ Whitehead et al.¹ subsequently noted that nonmicronized P administered orally to five postmenopausal women for 5 consecutive days resulted in peak P concentrations of 9.5 ng/ml. In these subjects, a sustained elevation of P was maintained for more than 96 hours after the 5-day loading dose.

Investigation of methods to increase the bioavailability of oral steroids resulted in experiments that indicate that a reduction in the particle size of P by micronization permits increased aqueous dissolution in the intestine and an increased plasma concentration of P in male rats.⁵ Subsequently, in preliminary human experiments, Morville et al.² reported mean peak P concentrations of 12.6 ng/ml after oral administration of micronized P dissolved in oil.

The present study evaluates the bioavailability and short-term toxicity of a simple oral prepara-

tion of micronized P. Gelatin capsules were chosen for simplicity and because minimal drug migration into the capsule shell was anticipated.⁹

After an overnight fast, a single oral dose of micronized P, 200 mg, prompted a significant increase in serum P concentrations in all patients

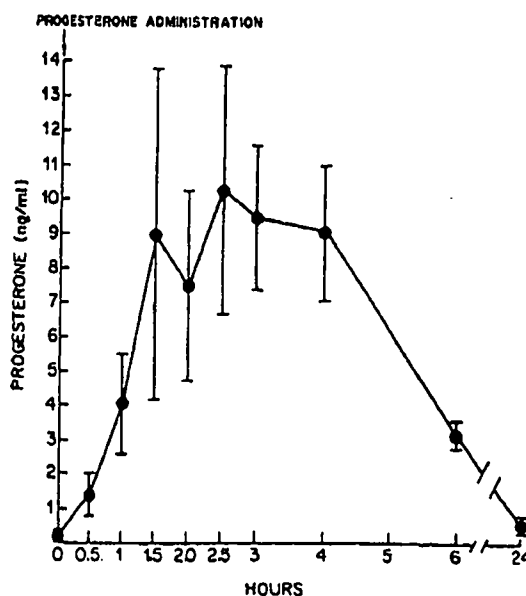


Figure 3
P concentrations (mean $\pm$ SEM) after oral micronized P, 200 mg at time 0.

by 2 hours. Mean peak concentrations (17.0 ± 4.9 ng/ml) appeared to be more than double those reported after 100 mg of nonmicronized P^{1, 5} and higher than 200 mg of micronized P in oil.² An accurate comparison of peak P concentrations after the various oral preparations is not possible because of the difference in RIA techniques employed.

P concentrations began to decline 4 hours after administration but remained significantly elevated for more than 6 hours, returning to baseline levels by 24 hours after administration. The time course of P absorption and metabolism is similar to that reported by Nillius and Johansson¹⁰ to occur after vaginal or rectal administration of nonmicronized P, 100 mg, in suppository form.

Wide interpatient variability in serum P concentrations was observed. This may be due to individual differences in the site and rate of absorption, clearance rates, and extent of absorption of P into fatty tissues. Such a variability in absorption could have significant impact on the clinical efficacy of oral P therapy.

Previous studies have demonstrated that P taken orally is physiologically active which produces a significant increase in tissue P concentrations in myometrium, endometrium, and breast^{2, 4} and induces histologic and biochemical changes in the endometrium after 10 days of administration in estrogen-primed postmenopausal women.⁶

Cortisol, aldosterone, and E₂ concentrations in these subjects did not change significantly over the course of the study, which confirms minimal conversion of P to these steroids, as reported previously. However, a significant positive correlation was found between baseline E₂ concentrations and the peak P levels achieved. Villanueva et al.¹¹ have demonstrated that the most rapid absorption and highest levels of P after vaginal application occurred in those postmenopausal women who were receiving estrogen. The authors hypothesized that anatomic and metabolic differences in the estrogen-treated group were responsible for improved P absorption.¹¹ It is unknown whether estrogen has a similar effect on the intestinal absorption and hepatic metabolism of P after oral administration.

No metabolic effects were seen in the parameters measured during the 24-hour period of observation in this study, and no major side effects were reported.

Our data clearly demonstrate that a single oral dose of 200 mg of micronized P provides P concentrations within or above the levels normally seen during the luteal phase. Further long-term studies are required to evaluate the clinical efficacy, side effects, and optimal dosage interval of prolonged oral administration of micronized P.

Acknowledgment. Micronized P was donated by The Upjohn Company, Kalamazoo, MI.

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Prediction of Stability in Pharmaceutical Preparations XVI: Kinetics of Hydrolysis of Canrenone and Lactonization of Canrenoic Acid

EDWARD R. GARRETT and CHONG MIN WON

Abstract □ The kinetics of the hydrolysis of the lactone canrenone, 3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionic acid γ -lactone, and the lactonization of its corresponding canrenoic acid salt, potassium 3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionate, were studied by spectrophotometrically analyzing the chloroform-extracted lactone from canrenoic acid solution. The log k -pH profiles in the pH range of 1-12 in various buffer solutions at 25.0, 37.5, 45.0, 60.0, 70.0, and 79.9° show that the kinetics of lactonization include hydrogen-ion attack on the undissociated canrenoic acid molecule and a pH-independent closure of the canrenoate anion. In addition to the hydroxide-ion-catalyzed hydrolysis of the lactone, general base-catalyzed hydrolysis in carbonate buffer solution was observed and attributed to carbonate-dianion attack. The rate constants, equilibrium constants, pK a' values, solubilities, and Arrhenius' parameters were obtained. Maximum concentrations of canrenoic acid salt to maintain elegant pharmaceutical preparations are given as a function of pH.

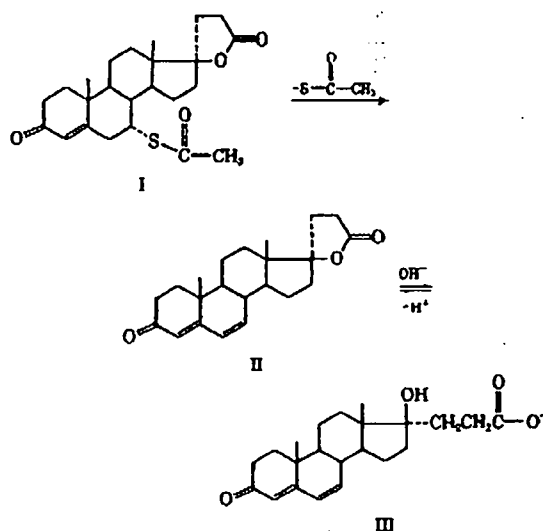
Keyphrases □ Canrenone, canrenoic acid—hydrolysis—lactonization kinetics, pH effect, appropriate pharmaceutical preparations □ Pharmacokinetics—canrenone hydrolysis, canrenoic acid lactonization, pH effect □ Hydrolysis—canrenone □ Lactonization—canrenoic acid □ UV spectrophotometry—analysis, canrenone and canrenoic acid

Spirolactone (I), 3-(3-oxo-7 α -acetylthio-17 β -hydroxy-4-androsten-17 α -yl)propionic acid γ -lactone, has been widely used in the treatment of edema that has not responded properly to treatment with conventional diuretics (1). Spirolactone exhibits a specific antagonism to the tendency of the adrenal steroid aldosterone to increase the reabsorption of sodium by reversibly competing with aldosterone at the receptor sites and thus modifying the sodium-retaining electrolyte excretion pattern, which is said to be the mechanism responsible for the production and maintenance of edema (2-4).

Canrenone (II), aldadiene [3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionic acid γ -lactone], may be the conjugated diene steroid found in the plasma after the oral administration of spironolactone (5). It may be formed by the elimination of the thioacetate group of spironolactone and may possess similar biological properties as an aldosterone antagonist (Scheme I).

As is common with γ -lactones, canrenone (II) is stable in acid and hydrolyzes in alkali to the corresponding canrenoic acid salt (III), potassium 3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionate. This study was undertaken to determine the quantitative transformations of canrenone in aqueous solution as a function of pH to supply the basic information necessary for preparing stable aqueous pharmaceutical preparations of canrenone and canrenoic acid.

In addition, it is important to know the case of the acid-lactone transformation to anticipate the species that may appear in the body under GI and other physiological conditions. As yet, it has not been clarified whether the lactone or the acid, or a metabolite derived from one or the other, is the pharmacologically active compound. The two species also might show different tendencies for protein binding and, hence, influence the distribution pattern of the drug in the



Scheme I

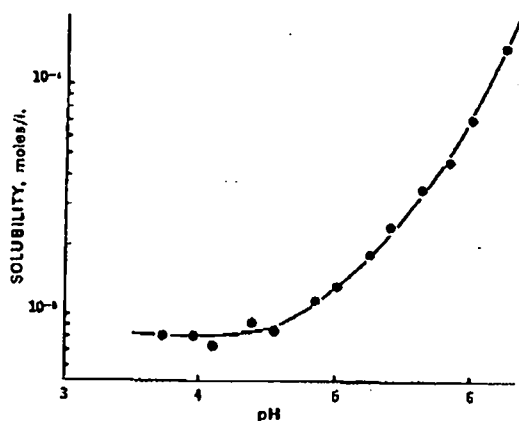


Figure 1—Semilogarithmic plot of the molar solubility of canrenoic acid as a function of pH at 25.0°.

body. Thus, this study on the interconversion of lactone and acid can serve as background for biopharmaceutical and pharmacokinetic studies of canrenone and canrenoic acid and as a basis for optimal design of appropriate dosage forms.

EXPERIMENTAL

Materials and Reagents—Canrenone (II), 3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionic acid γ -lactone, and the potassium salt of canrenoic acid, potassium canrenoate (III), potassium 3-(3-oxo-17 β -hydroxy-4,6-androstadien-17 α -yl)propionate, were used in this study¹. All other chemicals were of reagent grade.

Kinetic Procedures—A filtered aqueous solution of canrenone (about 9×10^{-3} M) was used as a stock solution for the kinetic studies on lactone hydrolysis. An aqueous stock solution of potassium canrenoate (about 4×10^{-2} M) was used for the kinetic studies on lactonization. The kinetic studies of hydrolysis and lactonization of canrenone and canrenoate anion, respectively, were made on final solutions in the concentration range of 1.0 – 3.6×10^{-3} M.

For slow reactions, i.e., half-lives in days, 5–20 ml. of stock buffer solutions and appropriate amounts of 1 M KCl to maintain an ionic strength of 0.1 (0.16 in phosphate buffer) were added to appropriately diluted volumes of the stock solutions. The first

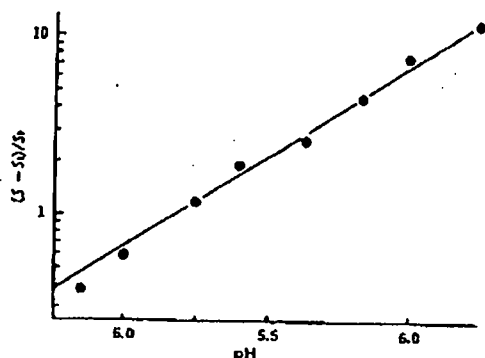


Figure 2—Semilogarithmic plot of the solubility function $(S - S_0)/S_0$ against pH at 25.0° in accordance with $\log(S - S_0)/S_0 = \text{pH} - \text{pKa}'$, where S is the total molar solubility of canrenoic acid and its anion at a given pH, and S_0 is the intrinsic solubility of the undissociated canrenoic acid. The slope is unity, and the intercept permits the calculation of pKa' as 5.20.

¹ Obtained from G. D. Searle & Co. as SC-9376 and SC-14266' respectively.

analyses of lactone concentration were made after the reacting solutions equilibrated in the temperature bath.

For the fast reactions, i.e., half-lives in hours or minutes, 99.0 ml. of appropriately diluted stock solutions of the reacting substances, adjusted to appropriate ionic strengths, was preheated in the temperature bath prior to the addition of 1.0 ml. of the acid or alkali used to accelerate the reaction. The bath temperatures were maintained within $\pm 0.1^\circ$ for 79.9° and within $\pm 0.05^\circ$ for all other temperatures.

The compositions of the buffer solutions at the various temperatures studied are given in Table I. Samples of the reacting solutions were taken at various times, cooled to room temperature, and analyzed for the lactone. The fast reactions were followed for 5–6 half-lives and the slow ones for at least 2–3 half-lives. The concentration of the lactone at infinite time was obtained after the reaction went 10 half-lives.

Assay Procedure—Cooled aliquots (4.00 ml.) of the reacting solutions were put into a 30-ml. separator, which contained 5.00 ml. of chloroform and 3.00 ml. of carbonate buffer ($[\text{CO}_3^{2-}] = 0.15$ M, $[\text{HCO}_3^-] = 0.05$ M), pH 10.4. The funnel was shaken vigorously for 1 min., the chloroform layer was separated, and the absorbance of the extracted lactone was read at 280 nm. with a slit width of 0.2 mm. against a buffer-saturated chloroform blank in a spectrophotometer². Within the significance of the spectral assay, all of the lactone was extracted into the chloroform layer under these conditions, and none could be spectrophotometrically observed in the aqueous layer. However, all of the potassium canrenoate remained in the aqueous layer under these conditions, and none could be spectrophotometrically observed in the chloroform layer.

Determination of Equilibrium Constants—The equilibrium constants were determined at different pH values at an ionic strength of 0.1 by analyzing the concentrations of the lactone and the acid in the sample after the reaction reached an equilibrium. Measurements also were made after the rate studies confirmed the achievement of this equilibrium. The assay procedure was used to determine the lactone concentration. The total acid and lactone concentration was obtained by partitioning both acid and lactone into 5.00 ml. of the chloroform layer by shaking 4.00 ml. of the sample solution together with the chloroform and 3.00 ml. of formate buffer ($[\text{HCOOH}] = 0.16$ M, $[\text{HCOONa}] = 0.04$ M), pH 3.0. The absorbance of the chloroform layer was read against a buffer-saturated chloroform blank at 280 nm. with a slit width of 0.2 mm. The molar absorptivity ($\epsilon = 29,000$) at the wavelength of maximum absorption (280 nm.) of the lactone canrenone and canrenoic acid in the chloroform was the same. The acid concentration was then calculated from the observed difference of the two assay procedures.

pKa' Determination—The pKa' (apparent pKa) of canrenoic acid (III) was determined by measuring the solubilities of the acid as a function of pH. A series of acetic acid-acetate, monohydrophosphate-dihydrophosphate solutions, ionic strength 0.1, ranging in pH values from 3.7 to 6.2, was prepared. Potassium canrenoate solution (1.00 ml. of 2.8×10^{-2} M) was added to 99.0 ml. of each buffer solution. This addition produced a precipitate of canrenoic acid whose solubility was exceeded at these pH values. The solutions were shaken at 25.0° in a mechanical shaker. After the solutions equilibrated, they were filtered. After adjustment of an aliquot with carbonate buffer to pH 10.4, as previously described for the lactone assay procedure, the resultant canrenoic acid solution was extracted with chloroform to remove the lactone; the separated buffered aqueous layers were spectrophotometrically assayed at λ_{max} 294 nm. ($\epsilon = 25,000$) at pH 10.4 for the sum of the canrenoic acid and its anion concentrations in the original solution at the various pH values. From these values, the total solubility of canrenoic acid and its anion could be calculated at the several pH values. The pKa' values at higher temperatures were determined from similar solubility determinations at the two pH values of 4.0 and 5.2 (acetate buffer). The intermittently shaken solutions were maintained in a constant-temperature water bath.

RESULTS AND DISCUSSION

Estimation of pKa' —The low solubility of canrenoic acid (III) in water necessitated the use of the solubility method for the

² Beckman DU.

Table I—Apparent First-Order Rate Constants, k^a , in sec^{-1} for the Achievement of Equilibrated Lactonization of Canrenoic Acid and Hydrolysis of Canrenoate as a Function of pH^b at Various Temperatures

Medium ^c	-25.0°		-37.5°		-45.0°	
	pH	k	pH	k	pH	k
(HCl)						
0.100	1.10	6.91×10^{-1}	—	—	—	—
0.070	1.23	4.77×10^{-1}	—	—	—	—
0.050	1.38	3.55×10^{-1}	—	—	—	—
0.040	1.47	2.80×10^{-1}	—	—	—	—
0.010	2.04	7.45×10^{-1}	2.04	1.62×10^{-1}	2.04	3.44×10^{-1}
[HCOOH] [NaOH]						
0.050 0.010	3.05	1.01×10^{-11}	3.00	2.00×10^{-11}	3.00	4.46×10^{-11}
[CH ₃ COOH][CH ₃ COONa]						
0.040 0.010	4.00	9.98×10^{-10}	4.00	2.65×10^{-10}	4.00	4.43×10^{-10}
0.010 0.040	5.23	1.08×10^{-10}	—	—	5.15	4.11×10^{-10}
[Na ₂ HPO ₄] [KH ₂ PO ₄]						
0.005 0.020	6.28	1.13×10^{-10}	—	—	—	—
0.016 0.004	—	—	7.42	5.94×10^{-10}	—	—
0.018 0.002	—	—	7.75	6.91×10^{-10}	—	—
[H ₂ BO ₃] [NaOH]						
0.010 0.003	8.80	7.73×10^{-10}	—	—	8.70	8.71×10^{-10}
[Na ₂ CO ₃] [NaHCO ₃]						
0.0020 0.0040	9.60	6.93×10^{-11}	9.65	2.50×10^{-10}	9.60	4.80×10^{-10}
0.0035 0.0045	—	—	10.00	5.20×10^{-10}	—	—
0.0060 0.0060	—	—	—	—	10.05	1.23×10^{-10}
0.0045 0.0035	—	—	10.15	7.70×10^{-10}	—	—
0.0053 0.0027	10.25	2.25×10^{-11}	10.30	9.10×10^{-11}	—	—
[NH ₄ CH ₃ COOH][NaOH]						
0.0104 0.0096	10.65	4.30×10^{-11}	—	—	—	—
[NaOH]						
0.005	11.6	5.43×10^{-11}	—	—	—	—
0.010	12.0	1.20×10^{-11}	—	—	—	—
0.015	12.1	1.89×10^{-11}	—	—	—	—
0.020	12.2	2.35×10^{-11}	—	—	—	—
[CH ₃ COOH][CH ₃ COONa]						
0.040 0.010	—	—	4.00	1.89×10^{-10}	—	—
0.010 0.015	—	—	4.80	3.28×10^{-10}	—	—
0.010 0.040	5.20	5.72×10^{-10}	5.20	1.10×10^{-10}	5.20	2.90×10^{-10}
[Na ₂ HPO ₄] [KH ₂ PO ₄]						
0.005 0.020	—	—	6.20	1.05×10^{-10}	6.20	2.75×10^{-10}
0.006 0.004	7.00	4.68×10^{-10}	7.00	9.60×10^{-11}	7.00	2.81×10^{-10}
0.016 0.004	7.40	6.86×10^{-10}	7.40	1.39×10^{-10}	7.40	3.91×10^{-10}
0.018 0.002	7.80	9.14×10^{-10}	7.80	2.05×10^{-10}	7.80	6.56×10^{-10}
[H ₂ BO ₃] [NaOH]						
0.010 0.003	—	—	8.70	8.75×10^{-11}	—	—

^a The rate constants are the sums of the hydrolysis and the lactonization rate constants. They are the values obtained by extrapolation to zero buffer concentration whenever a buffer catalysis was observed. ^b The pH values in HCl and NaOH were calculated from $\text{pH} = -\log \gamma_{\text{HCl}}(\text{HCl})$ and $\text{pH} = \text{pKw} - \text{pOH}$, where $\text{pOH} = -\log \gamma_{\text{NaOH}}(\text{NaOH})$ where the activity coefficients, γ_{HCl} and γ_{NaOH} , and pKw values were obtained from the literature (10). All other pH values were measured by a Radiometer pH meter equipped with a Corning semimicro combination electrode (temperature range -5 ~ 100°). The pH meter was standardized with standard buffers (Sargent-Welch) of pH 4.0, 7.0, and 10.0 at the same temperature. ^c KCl was added to maintain the ionic strength, μ , at 0.1 (0.16 in phosphate buffer). ^d The 10% for multiples of the stated buffer concentration were: 1X, 0.98; 2X, 0.96; 3X, 1.10; and 4X, 1.60. ^e The 10% for multiples of the stated buffer concentration were: 1X, 1.07; 2X, 1.07; 3X, 1.14; and 4X, 1.19. ^f The 10% for multiples of the stated buffer concentration were: 1X, 1.13; 2X, 1.46; 3X, 1.68; and 4X, 1.92. ^g The 10% for multiples of the stated buffer concentration were: 1X, 7.14; 1.5X, 7.33; 3.5X, 7.90; and 5.0X, 8.04. ^h The 10% for multiples of the stated buffer concentration were: 0.75X, 2.32; 1.13X, 2.68; 2.64X, 3.02; and 3.77X, 3.19. ⁱ The 10% for multiples of the stated buffer concentration were: 1X, 4.77; 2X, 6.17; 3X, 7.00; and 4X, 7.45. ^j The 10% for multiples of the stated buffer concentration were: 1X, 2.84; 2X, 3.03; 3X, 3.34; and 4X, 3.23. ^k The 10% for multiples of the stated buffer concentration were: 1X, 5.94; 1.5X, 5.94; 2.0X, 5.94; and 2.5X, 5.94. ^l The 10% for multiples of the stated buffer concentration were: 1X, 6.91; 1.5X, 6.91; 2.0X, 6.91; and 2.5X, 6.91. ^m The 10% for multiples of the stated buffer concentration were: 1X, 5.48; 2X, 5.97; 3X, 6.17; and 4X, 6.66. ⁿ The 10% for multiples of the stated buffer concentration were: 1X, 7.63; 2X, 8.44; 3X, 9.29; and 4X, 9.59. ^o The 10% for multiples of the stated buffer concentration were: 1X, 9.44; 2X, 10.3; 3X, 11.0; and 4X, 11.1. ^p The 10% for multiples of the stated buffer concentration were: 1.0X, 4.61; 1.5X, 5.22; 2.0X, 5.49; and 2.5X, 5.57. ^q The 10% for multiples of the stated buffer concentration were: 1X, 4.11; 2X, 4.11; 3X, 4.11; and 4X, 4.11. ^r The 10% for multiples of the stated buffer concentration were: 1.0X, 5.20; 1.5X, 5.43; 3.5X, 6.14; and 5.0X, 6.95. ^s The 10% for multiples of the stated buffer concentration were: 1X, 1.23; 2X, 1.23; 3X, 1.23; and 4X, 1.23. ^t The 10% for multiples of the stated buffer concentration were: 1.0X, 1.39; 1.5X, 1.39; 2.0X, 1.54; and 2.5X, 1.51. ^u The 10% for multiples of the stated buffer concentration were: 1.0X, 1.34; 1.5X, 1.28; 2.0X, 1.47; and 2.5X, 1.47. ^v The 10% for multiples of the stated buffer concentration were: 1.0X, 2.38; 1.5X, 2.38; 2.0X, 2.62; and 2.5X, 2.71. ^w The 10% for multiples of the stated buffer concentration were: 1.0X, 2.65; 1.5X, 2.66; 2.0X, 2.99; and 2.5X, 3.10. ^x The 10% for multiples of the stated buffer concentration were: 1.0X, 2.84; 1.5X, 2.84; 2.0X, 2.81; and 2.5X, 2.77.

determination of the ionization constant. If S_0 is the intrinsic solubility of the undissociated acid, $[HA]$, the ionization constant:

$$K_a' = [H^+][A^-]/[HA] \quad (\text{Eq. 1})$$

can be expressed in terms of the solubilities (6) as:

$$K_a' = [H^+](S - S_0)/S_0 \quad (\text{Eq. 2})$$

$$\log [(S - S_0)/S_0] = \text{pH} - \text{pK}_a' \quad (\text{Eq. 3})$$

where S is the total solubility of the undissociated canrenoic acid in equilibrium with its anion at a given pH.

When the logarithm of $(S - S_0)/S_0$ is plotted against pH, a straight line is produced with a slope of unity and the negative intercept of pK_a' . The analytical procedure was presumed to remove any lactone formed during the equilibrium process, and lactonization was presumed to be a slower process than the equilibration between the solid canrenoic acid and its saturated solution. The solid phase of the equilibrated mixture was confirmed to be canrenoic acid by

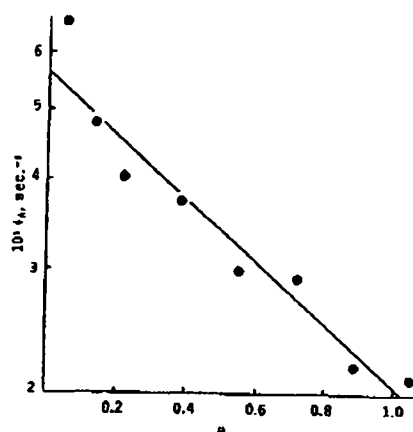


Figure 3—Semilogarithmic plot of the apparent first-order rate constants in the hydrolysis of canrenone as a function of ionic strength, μ , in carbonate buffer (pH 10.5) at 25.0°.

the fact that it was instantaneously soluble in mildly alkaline aqueous solution.

As would be expected from the relations of Eqs. 2 and 3, the solubility approaches the constant value S_0 as the pH decreases (Fig. 1). The plot of $\log [(S - S_0)/S_0]$ versus pH is shown in Fig. 2. The intercept estimates a pK_a' value of 5.20 at 25.0° (Eq. 3). No pK_a' dependence on temperature was observed within experimental error. The experimental pK_a' values at the various temperatures are: 25.0°, 5.20; 37.5°, 5.21; 45.0°, 5.25; and 60.0°, 5.02.

Effect of Ionic Strength on Hydrolysis Rate—The hydrolysis of canrenone in carbonate buffer of pH 10.5 was tested for a salt effect with KCl up to ionic strengths of 1.25. A negative salt effect was observed (Fig. 3) since the logarithms of the hydrolysis rate constant significantly and linearly decreased with ionic strength. At higher ionic strengths (7, 8), the activity coefficient of an ion in aqueous solution at 25° adheres to:

$$\log \gamma_i = -0.509z_i^2 \sqrt{\mu} + b_i \mu \quad (\text{Eq. 4})$$

where z_i is the charge on species i . It can be shown by transition-

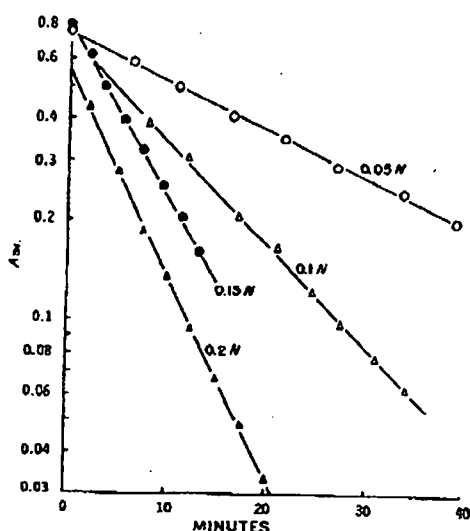


Figure 4—Typical first-order plots for hydrolysis of canrenone in various NaOH concentrations at 25.0°. The absorbance values, A , of the unreacted lactone were measured at 284 nm. in the chloroform extract.

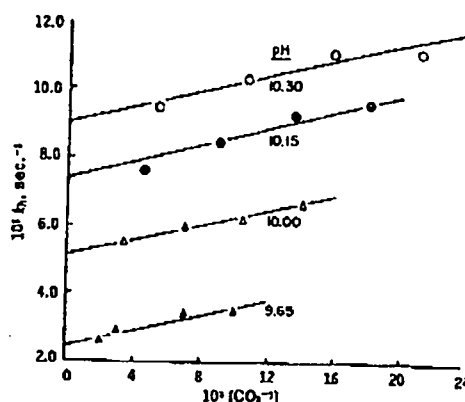


Figure 5—Apparent first-order rate constants in hydrolysis of canrenone in carbonate buffer at 37.5° as a function of pH and the concentration of buffer anion, $[CO_3^{2-}]$.

state theory (9) that:

$$\log k = \log k_0 + (b_0 + b_i - b^\ddagger)\mu \quad (\text{Eq. 5})$$

where b_i is the coefficient of ionic strength for hydroxide ion, b_0 is for the lactone, and $b^\ddagger$ is for the activated complex, when k_0 is the rate constant at zero ionic strength. The conformity of the data of Fig. 3 to Eq. 5 indicates that the ionic concentration has a greater effect on the activated complex formed from hydroxide ion and lactone than it does on the two separated species. Of course, the possibility does exist that the activity of the lactone itself is decreased with increased ionic strength.

Rate Constants—The apparent first-order rate constants (Table I) were estimated from the slopes of appropriately plotted experimental data in accordance with:

$$\log [A_\infty - A] = -kt/2.303 + \log [A_\infty - A_0] \quad (\text{Eq. 6})$$

or by the method of least squares using the Wang 700 calculator. There was no significant difference between the statistical and graphical methods. The A_∞ values were the final absorbances of the reaction, the A_0 values were the initial readings, and the absorbance values A were read at any given time t . At any constant pH, semilogarithmic plots in accordance with Eq. 6 increased or decreased linearly with time to provide confidence in k as an apparent first-order rate constant. Typical first-order plots for various NaOH concentrations are given in Fig. 4.

The separation of the apparent first-order lactonization (k_l) and hydrolysis (k_h) rate constants was achieved with the use of the equilibrium constants, since the apparent first-order rate constant for the achievement of an equilibrium is the sum of the forward and backward rate constants (Eq. 7) and the equilibrium constant is the quotient (Eq. 8):

$$k = k_h + k_l \quad (\text{Eq. 7})$$

$$K = k_h/k_l \quad (\text{Eq. 8})$$

Thus, k_h and k_l can be obtained from the experimental k and K values obtained at a given pH value.

General Acid-Base Catalysis—The rate constants were independent of buffer concentration at constant pH and ionic strength for formate and borate buffers and gave no evidence of general acid-base catalysis. The buffer catalysis by acetate and phosphate buffers was insignificant. However, significant buffer catalysis was observed with carbonate and glycine buffers in the hydrolysis of canrenone. The rate constants for the reactions studied in different buffer concentrations are given in the footnotes of Table I.

The possible catalytic contributions to the apparent first-order rate constant in the carbonate buffer region where only complete hydrolysis of the lactone was observed, on the assumption of no water attack, may be expressed by:

$$k_h = k_{OH}[OH^-] + k_{HCO_3}[HCO_3^-] + k_{CO_3}[CO_3^{2-}] \quad (\text{Eq. 9})$$

here k_1 is the apparent first-order rate constant for the hydrolysis of the lactone canrenone, and k_{OH^-} , $k_{HCO_3^-}$, and $k_{CO_3^{2-}}$ are the respective catalytic rate constants. Since, when $K_{HCO_3^-}$ is the dissociation constant of HCO_3^- :

$$[HCO_3^-]/[CO_3^{2-}] = [H^+]/K_{HCO_3^-} \quad (\text{Eq. 10})$$

$$= k_{OH^-}[OH^-] + (k_{HCO_3^-}[H^+]/K_{HCO_3^-} + k_{CO_3^{2-}}[CO_3^{2-}]) \quad (\text{Eq. 11})$$

$$k_1 = k_{OH^-}[OH^-] + (k_{HCO_3^-} + k_{CO_3^{2-}}K_{HCO_3^-}/[H^+])[HCO_3^-] \quad (\text{Eq. 12})$$

Plots of k_1 against $[HCO_3^-]$ or $[CO_3^{2-}]$ at constant $[H^+]$ should be linear and of positive slopes with intercept values of $(k_{OH^-}[H^-])$. When the slopes, S_1 , of Eq. 11:

$$S_1 = k_{HCO_3^-}[H^+]/K_{HCO_3^-} + k_{CO_3^{2-}} \quad (\text{Eq. 13})$$

is plotted against the hydrogen-ion activity, the resultant slope $k_{HCO_3^-}/K_{HCO_3^-}$ and the resultant intercept is $k_{CO_3^{2-}}$. Similarly, from the slopes, S_2 , of Eq. 12 against $1/[H^+]$:

$$S_2 = k_{HCO_3^-} + k_{CO_3^{2-}}K_{HCO_3^-}/[H^+] \quad (\text{Eq. 14})$$

$k_{CO_3^{2-}}$ and $k_{HCO_3^-}$ can be evaluated separately when $K_{HCO_3^-}$ is known. The apparent rate constant, k_1 , was plotted against $[CO_3^{2-}]$ (Fig. 5) and $[HCO_3^-]$ (Fig. 6). The fact that the slopes (S_1) in Fig. 5 are independent of pH within experimental error indicates the non-variation of $(k_{HCO_3^-}[H^+]/K_{HCO_3^-} + k_{CO_3^{2-}})$ with $[H^+]$ and, thus, the lack of a catalytic rate constant, $k_{HCO_3^-}$, in carbonate buffer-catalyzed hydrolysis of the lactone. The catalytic rate constant, $k_{CO_3^{2-}}$, was $1.1 \times 10^{-3} \text{ sec}^{-1}$ at 37.5° . The identical value of $k_{CO_3^{2-}}$ can be obtained from the slope of plots of the slopes (S_2) of Fig. 6 against $1/[H^+]$ in accordance with Eq. 12. The intercept of zero confirms the fact that $k_{HCO_3^-} \sim 0$.

The intercepts of the plots in Figs. 5 and 6 vary and are consistent with hydroxyl-ion catalysis; i.e., the intercepts $(k_{OH^-}[OH^-])$ versus $[OH^-]$ give a linear plot of slope k_{OH^-} . The line passes through zero and confirms the fact that there is no water hydrolysis. The k_{OH^-} value estimated by such a method was 0.21 sec^{-1} at 37.5° . The rate constants were extrapolated to zero buffer concentrations, and the determined $k_{OH^-}[OH^-]$ values were used in the log k -pH profiles.

Specific Acid-Base Catalysis.—Plots of the apparent first-order rate constants against HCl and NaOH (Fig. 7) in strongly acidic and alkaline regions were linear and with zero intercept in accordance with:

$$k_1 = k_{HCl}[HCl] \quad (\text{Eq. 15})$$

$$k_1 = k_{NaOH}[NaOH] \quad (\text{Eq. 16})$$

In these highly acidic regions, all canrenonic acid was converted to the lactone canrenone within the precision of the experimental method. In these highly alkaline regions, all of the lactone canrenone

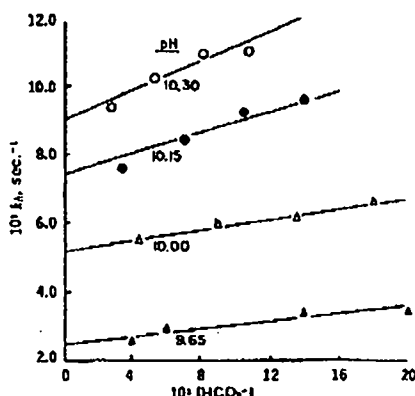


Figure 6—Apparent first-order rate constants in hydrolysis of canrenone in carbonate buffer at 37.5° as a function of pH and the concentration of buffer acid, $[HCO_3^-]$.

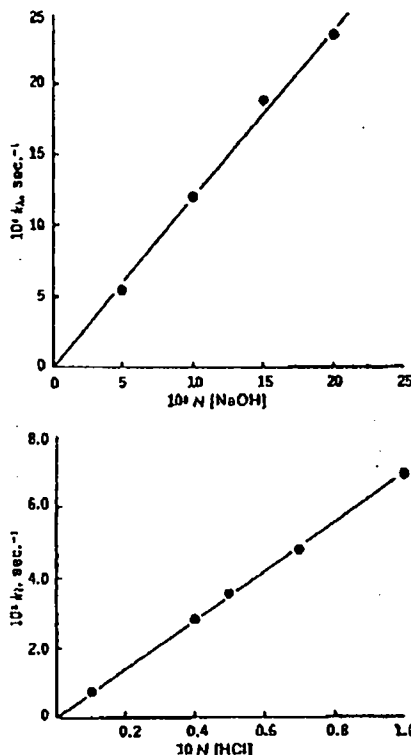


Figure 7—Apparent first-order rate constant dependence on $[NaOH]$ for hydrolysis of canrenone (top) and on $[HCl]$ for lactonization of canrenonic acid (bottom) at 25.0° .

none was converted to the canrenonic acid within the precision of the experimental method. Both lines (Fig. 7) had zero intercepts to indicate the lack of water reaction.

Equilibrium Constants.—The equilibrium constants at any pH value can be defined as the ratio of apparent first-order hydrolysis

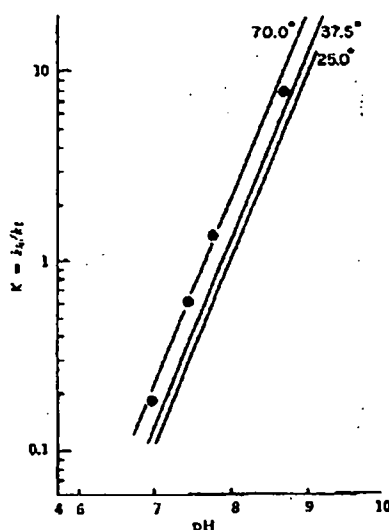


Figure 8—Semilogarithmic plot of equilibrium constants, $K (=k_1/k_2)$, as a function of pH. The drawn lines at 25.0 and 37.5° were calculated from the microscopic rate constants with the use of Arrhenius plots.

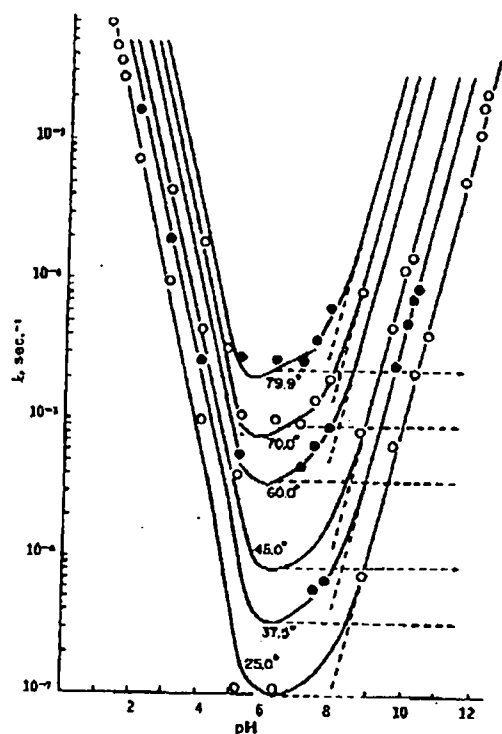


Figure 9—Log k -pH profile for the lactonization of canrenic acid and hydrolysis of canrenone at 25.0, 37.5, 45.0, 60.0, 70.0, and 79.9°. The drawn lines were calculated from the derived microscopic rate constants. The dashed lines were the separated log k -pH profiles for lactonization and hydrolysis.

and lactonization constants. Consequently, it is a function of hydrogen- and hydroxyl-ion activity. The experimental equilibrium constants ($K = k_1/k_2$) at 70.0° are plotted semilogarithmically in Fig. 8. Experimental values of K obtained at 79.9° were not significantly different.

Log k -pH Profile—The log k -pH profiles were constructed from the first-order rate constants (Table I) and pH values at 25.0, 37.5, 45.0, 60.0, 70.0, and 79.9°. The sum of the rate constants of the lactonization and the hydrolysis as a function of pH are plotted in Fig. 9. The separated log k -pH profiles for lactonization and hydrolysis are shown in this figure by the dashed lines. The pH values of the strongly acidic or strongly alkaline solutions were calculated from:

$$\text{pH} = -\log \gamma_{\text{HCl}}[\text{HCl}] \quad (\text{Eq. 17})$$

$$\text{pH} = \text{pKw} - \text{pOH} = \text{pKw} + \log \gamma_{\text{NaOH}}[\text{NaOH}] \quad (\text{Eq. 18})$$

where pKw and the activity coefficients, γ_{HCl} and γ_{NaOH} , were obtained from the data in the literature (10).

The logarithmic transformations of Eqs. 15 and 16 are:

$$\log k_1 = \log k_{\text{H}} - \text{pH} \quad (\text{Eq. 19})$$

$$\log k_2 = \log k_{\text{OH}} - \text{pKw} + \text{pH} \quad (\text{Eq. 20})$$

where the specific bimolecular rate constants, k_{H} and k_{OH} , are now defined with respect to hydrogen- and hydroxide-ion activities rather than their respective molarities, as were the k_{HCl} and k_{NaOH} of Eqs. 19 and 20. The separated k_1 and k_2 values were calculated on the basis of the observed overall first-order rate constants of change (Table I and Fig. 9) and the determined equilibrium constants (Fig. 8). The bases for this separation were given previously in Eqs. 7 and 8.

The log k_1 decreases with decreased pH with slope of unity in accordance with Eq. 20 (Fig. 9). The apparent lactonization rate constant conforms to the dependency of Eq. 21 below a pH of 3

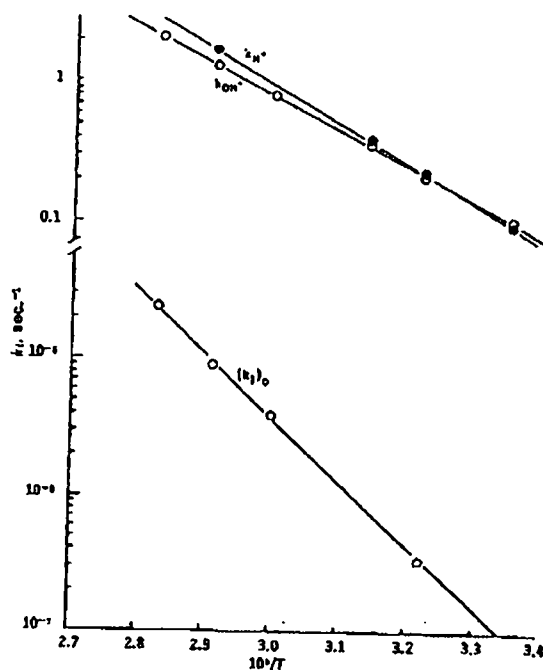


Figure 10—Arrhenius plots of the microscopic rate constants for lactonization of canrenic acid, k_{H} and $(k_1)_0$, and hydrolysis of canrenone, k_{OH} .

where essentially all the acid molecules are undissociated. The apparent first-order lactonization rate constant tends to be independent of pH as the fraction of dissociated acid present increases (as pH tends to exceed the pK_a). This indicates a pH-independent lactonization of the anion. Hence, the overall lactonization can be formulated as:

$$k_1 = k_{\text{H}}[\text{H}^+]/f_{\text{HA}} + (k_1)_0 f_{\text{A}} = k_{\text{H}}[\text{H}^+]/([\text{H}^+] + K_a) + (k_1)_0 K_a/([\text{H}^+] + K_a) \quad (\text{Eq. 21})$$

where f_{HA} and f_{A} are the fractions of undissociated and dissociated acids, respectively (11), and $(k_1)_0$ is the rate constant for a pH-independent lactonization of the anion. The solid drawn lines in the log k -pH profile (Fig. 9) were derived from the sums of the first-order rate constants of Eqs. 20 and 21.

The fact that a plot of $\log K$ versus pH (Fig. 8) is linear and of positive slope of unity in the pH regions where the amounts of canrenone and canrenic acid assayed were significant indicates that:

$$K = k_1/k_2 = k_{\text{OH}}[\text{OH}^-]/(k_1)_0 \quad (\text{Eq. 22})$$

Table II—Microscopic Rate Constants^a, Arrhenius Parameters^b, and Entropies of Activation^c for Hydrolysis of Canrenone and Lactonization of Canrenic Acid

Temperature	k_{H} , sec. ⁻¹	k_{OH} , sec. ⁻¹	$10^4(k_1)_0$, sec. ⁻¹
25.0°	0.10	0.11	—
37.5°	0.24	0.21	34.0
45.0°	0.40	0.36	—
60.0°	—	0.80	400
70.0°	1.70	1.32	900
79.9°	—	2.12	2400
E_a , kcal.	12.84	11.42	21.23
$\log A$	8.40	7.38	8.50
$\Delta S^\ddagger$, e.u.	-20.0	-24.7	-19.5

^a Where $k_2 = k_{\text{OH}}[\text{OH}^-]$, $k_1 = k_{\text{H}}[\text{H}^+]/f_{\text{HA}} + (k_1)_0 f_{\text{A}}$. ^b Where $\log k = -E_a/2.303RT + \log A$. ^c Where $\Delta S^\ddagger/R = 2.303(\log A - \log(kT/h)) - 1$.

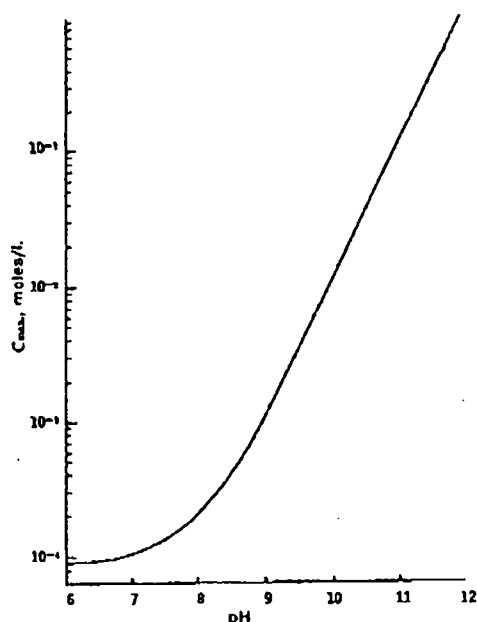


Figure 11—Semilogarithmic plot of the maximum molar concentration, C_{max} , of stabilizable potassium canrenoate in aqueous preparations as a function of pH at 25.0°.

so that:

$$\log K = \log [k_{OH}/(k_1)_0] - pK_w + pH \quad (\text{Eq. 23})$$

where the intercept $\log [k_{OH}/(k_1)_0] - pK_w = 7.96$ at 25.0°. Thus, in the pH region where significant amounts of both species are present in equilibrium, the hydrolysis rate must be hydroxide ion dependent and the lactonization rate must be pH independent.

If this was not the case and if hydrolysis was $[OH^-]$ dependent and lactonization was $[H^+]$ dependent:

$$K = k_1/k_2 = k_{OH}[OH^-]/k_2[H^+] \quad (\text{Eq. 24})$$

It would follow that:

$$\log K = \log (k_{OH}K_w/k_2) + 2pH \quad (\text{Eq. 25})$$

and the plot of Fig. 8 would be linear with positive slope of 2, which is not the case.

If lactonization of undissociated acid was pH dependent and that of dissociated acid was pH independent, and hydrolysis was hydrogen-ion as well as hydroxyl-ion catalyzed, then:

$$K = k_1/k_2 = (k_2[H^+] + k_{OH}[OH^-]) / (k_1[H^+]f_{HA} + (k_1)_0f_A) \quad (\text{Eq. 26})$$

Thus, it would follow that at high alkalinity when $[H^+]$ is negligible, the $\log K$ is the function of pH as given in Eq. 23 and justified by the plot of Fig. 8. This would also be true in the neutral pH region of Fig. 8 if $k_1[H^+]f_{HA}$ and $k_2[H^+]$ were negligible. At higher acidity, i.e., where $[H^+] \gg [OH^-]$, it would follow that $K = k_1/k_2$ so that the equilibrium constant would tend to become invariant with pH at low pH values and the plot of $\log K$ versus pH of Fig. 8 would tend to flatten at lower pH values. This could not be concluded within the experimental error. Although the acid-catalyzed hydrolysis of γ -lactones was observed in the hydrolysis of γ -butyrolactone (12), γ -substituted γ -lactones (13), and pilocarpine (14), one could not conclude that it was significant in the case of canrenone. At least the acid-catalyzed hydrolysis rate cannot be any more than 5% as large as the acid-catalyzed lactonization rate, i.e., the equilibrium constant in highly acidic solution cannot be greater than 0.05.

Microscopic Rate Constants—The values of the microscopic rate constants, k_2 and k_{OH} , at all temperatures were estimated

directly from the best fit of the $\log k$ -pH profile (Fig. 9) in low and high pH regions, respectively. The bimolecular rate constants of pH-independent lactonization of the dissociated acid were estimated from the best fit of the profile in the pH 7-8 region where essentially all the acid is in the dissociated form. The microscopic rate constants at different temperatures are given in Table II. The solid lines in the $\log k$ -pH profile (Fig. 9) were calculated from the k_i values in Table II.

Rate Dependency on Temperature—The Arrhenius equation gives the quantitative relation of rate constants and temperature. The logarithmic form of the equation is:

$$\log k = -E_a/2.303RT + \log A \quad (\text{Eq. 27})$$

where E_a is the activation energy, and A is the frequency factor. The Arrhenius plots of the logarithms of the various catalytic rate constants are shown in Fig. 10 for: k_{OH} , the specific base catalytic hydrolysis rate constant; k_2 , the specific acid catalytic lactonization rate constant; and $(k_1)_0$, the pH-independent lactonization rate constant of the dissociated acid. The Arrhenius parameters were calculated from the plots in accordance with Eq. 27. The values of E_a , $\log A$, and $\Delta S^\ddagger$ (entropy of activation) are included in Table II.

Since the rate of achievement of an equilibrium in the neutral pH regions at lower temperatures is an extremely slow process, the equilibrium constants at lower temperatures were calculated from the ratio of Eqs. 20 and 21. The microscopic rate constants given in these equations were based on the values of the constants estimated from the use of their Arrhenius parameters (Eq. 27 and Table II). Similar plots of such calculated equilibrium constants as a function of pH are given in Fig. 8 for 25.0 and 37.5°.

Mechanism for Hydrolysis of Canrenone and Lactonization of Canrenonic Acid—Long and Friedman (15), by using ^{18}O -labeled water, showed that acyl oxygen scission of a γ -lactone occurs in both the acid- and base-catalyzed reactions and that the hydrolysis passes through an intermediate with enough lifetime to allow

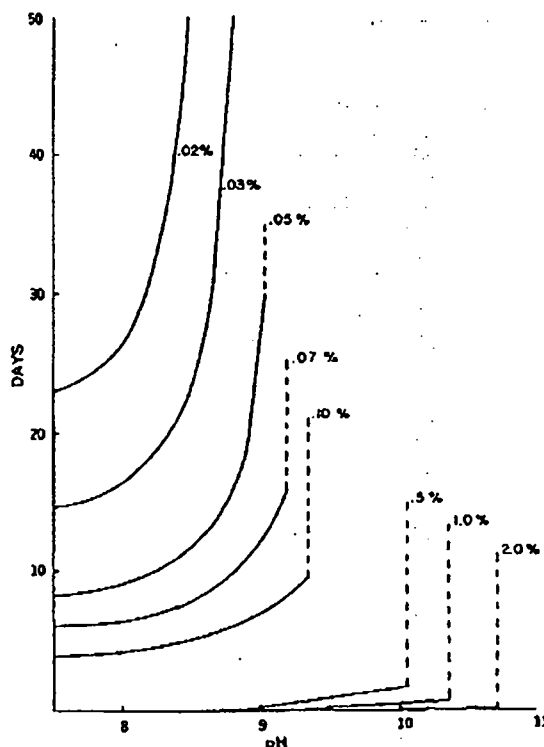
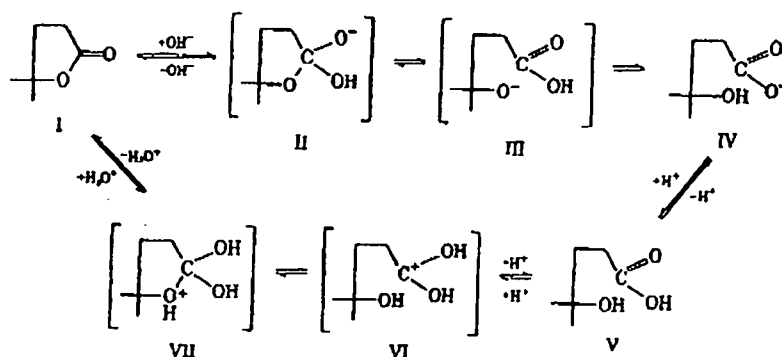


Figure 12—Time required to start forming precipitates of canrenone in aqueous preparations of potassium canrenoate as a function of pH and concentration of potassium canrenoate at 25.0°.

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Scheme II

appreciable oxygen exchange, such as that found in the hydrolysis of simple esters. Long *et al.* (12) also studied the kinetics of the acid-catalyzed hydrolysis of γ -butyrolactone in strongly acidic solutions and found that the rate of hydrolysis is proportional to the concentration of hydrogen ion, but the rate of the lactonization is proportional to h , (Hammett's acidity function). This indicated that the activated complex consisted not only of a molecule of lactone and hydrogen ion but also of a water molecule [Hammett-Zucker hypothesis (16)].

Scheme II is consistent with these mechanisms for lactonization and hydrolysis of γ -lactones. The sequence I $\rightarrow$ IV explains the base-catalyzed hydrolysis of canrenone and is consistent with a rate dependency of the apparent first-order rate constant on k_{OH^-} . It is also consistent with the pH-independent lactonization of the canrenoic acid monoanion, which may be initiated by an intramolecular general base attack on the alcoholic group (IV $\rightarrow$ III) (possibly mediated by a molecule of water), which then results in the intramolecular nucleophilic attack of the resultant alcoholate anion on the carbonyl carbon to produce the common activated complex, II.

The sequence V $\rightarrow$ VI $\rightarrow$ VII $\rightarrow$ I explains the acid-catalyzed lactonization of undissociated canrenoic acid and is consistent with a rate dependency of the apparent first-order rate constant on k_{H^+}/r_{HA} . The possible reverse reaction, the acid-catalyzed hydrolysis of canrenone, does not appear to be large enough to affect significantly the equilibrium for all practical purposes. This was also true for the acid-catalyzed hydrolysis of pilocarpine (14), although the acid-catalyzed hydrolysis of γ -butyrolactone was significant (12). Bruce and Pandit (17) showed that alkyl substitutions on the carbons of structures amenable to intramolecular reaction also favor the cyclization.

Prediction of Stability—The pH-independent lactonization occurs in the alkaline region at all pH values, as shown in the log k -pH profile (Fig. 9). The extent of its formation is a function of its equilibrium with the hydroxide-ion-catalyzed hydrolysis. The lactone canrenone precipitates when the concentration of the lactone is in equilibrium at a given pH value exceeds its solubility. Thus, the pharmaceutical stability of the canrenoic acid salt is a function of the solubility of the lactone and the pH-dependent thermodynamic equilibrium of the reversible lactonization and hydrolysis processes. The solubilities of the lactone, canrenone (S_L), in water were found to be: 25.0°, 9.4×10^{-5} M; 37.5°, 7.79×10^{-5} M; and 60.0°, 2.43×10^{-5} M. Substitution of the ratio of canrenoate anion (at pH values much higher than 5.2 where the acid concentration is negligible) to canrenone concentrations for the equilibrium constant K in Eq. 23 results in:

$$\log K = \log \left(\frac{[\text{canrenoate}]}{[\text{canrenone}]} \right) = \log \left(\frac{C_{\text{max}} - S_L}{S_L} \right) \\ = \log \left(\frac{C_{\text{max}} - 9.4 \times 10^{-5}}{9.4 \times 10^{-5}} \right) = \text{pH} - 7.96 \quad (\text{Eq. 28})$$

where C_{max} , in moles/l. is the maximum concentration of canrenoic acid salt that can be prepared at a given pH in solution at 25.0° and yet will not give an ultimate precipitate of equilibrated canrenone. A plot of C_{max} , calculated from Eq. 28 for various pH values is given in Fig. 11 and clearly shows the maximum concen-

trations of canrenoate solution that can be prepared at any specific pH value to maintain a true solution at 25.0°.

The time required to reach that point where the lactone will precipitate from a canrenoic acid salt solution is a function of the concentration of the canrenoic acid salt as well as the pH, since both the rate and equilibrium constants are pH dependent. The lactone concentration, L_{eq} , in moles/l. at equilibrium may be expressed by:

$$L_{\text{eq}} = C_0 / (1 + K) \quad (\text{Eq. 29})$$

where C_0 is the initial concentration in moles/l. of the canrenoic acid salt and $K = (C_0 - L_{\text{eq}}) / L_{\text{eq}}$. The apparent first-order transformation of canrenoate anion to the lactone, canrenone, at a constant pH may be expressed by:

$$\log (C - C_{\text{eq}}) = \log (C_0 - C_{\text{eq}}) - kt / 2.303 \quad (\text{Eq. 30})$$

where C is the concentration of nonlactonized canrenoic acid or anion at any time t , and k is the apparent first-order rate constant (Table I) to achieve an equilibrium. If the concentration, C , of canrenoate is limited so that precipitates of the lactone cannot occur, then:

$$C = C_0 - S_L \quad (\text{Eq. 31})$$

and, from Eq. 29:

$$C_{\text{eq}} = KL_{\text{eq}} = KC_0 / (1 + K) \quad (\text{Eq. 32})$$

Thus, substitution of Eqs. 31 and 32 into Eq. 30 and subsequent rearrangement result in

$$t_{SL} = (2.303/k) \log [C_0 / \{C_0 - S_L(1 + K)\}] \quad (\text{Eq. 33})$$

and the time, t_{SL} , when the lactone, canrenone, of solubility S_L precipitates from a solution of C_0 (moles/l.) concentration may be calculated at any temperature when the apparent first-order rate constant, k (Table I), for the achievement of the equilibrium and the equilibrium constant, K (Fig. 8), are known. These times, t_{SL} , required for precipitation of the lactone for various concentrations of the canrenoic acid salt are given in Fig. 12. Even a dilute solution of potassium canrenoate (0.02%) at pH 8.0 gives a precipitate in less than a month. Thus, if it is still desirable to have a soluble preparation of potassium canrenoate, either adjuncts should be added to increase the hydrolysis rate of the lactone without affecting the rate of lactonization or mixed solvents should be considered so that a small amount of lactone in the equilibrium will be completely solubilized.

Biopharmaceutical Aspects of Spironolactone, Canrenone, and Canrenoic Acid—Gantl *et al.* (18) reported originally that the addition of polysorbate 80 enhanced the absorption of spironolactone. Subsequently, they revised (19) their conclusion to state that the addition of polysorbate 80 does not contribute directly to the previously observed increased absorption. This revision was due to their observation that the smaller particle size of spironolactone was preserved when the drug was encapsulated with the surfactant; thus they attributed the improved absorption to the

particle size rather than to the addition of the surfactant. The smaller particle-sized spironolactone was as well absorbed as the one with polysorbate 80. Their observation was supported by several clinical and blood level studies (20-22). Venning (23) argued against this dependence of absorption on particle size by claiming that blood level studies demonstrated that conventional tablets with an average particle size of $1\ \mu$ were as poorly absorbed as those of $10\ \mu$. He concluded that "absorption of spironolactone is dependent upon some more complex function of the physical state within the finished tablets." The fact of the matter was that these two sets of tablets had different formulation factors. However, dissolution studies by Levy (24) showed that the dissolution rates were faster with the more readily absorbed new tablet formulations than with the old ones. He assigned this discrepancy to the dissolution rate-limited absorption of spironolactone.

If canrenoic acid salt were administered orally, the drug would be transformed readily to its undissociated form and undoubtedly precipitate as such in the stomach. Lactonization would be anticipated to occur prior to absorption at the pH of the stomach (pH 1) since the half-life of the lactonization is only 30 sec. The overall absorption process should be limited by the rate of dissolution and the intrinsic solubility of the canrenoic acid (5.7 mcg./ml. at pH 7.5). Certainly, the oral administration of the canrenoic acid salt may give more rapid dissolution (25), wherein it would be precipitated as a fine, readily redissolvable particle. Thus, this could serve as an alternative method of orally administering the lactone. In the less acidic conditions of the intestine, canrenoic acid would be more soluble and the absorption process would be facilitated if the dissolution is rate determining. Since the rate of lactonization is much slower at pH 5.3 at the absorption surface of the intestine (half-life 12 days), the major portion of the drug would be absorbed as the acid species rather than as the lactone. Once the drug is absorbed, either as canrenoic acid or as canrenone, it should remain as it is in the body fluids (hydrolysis or lactonization is slow at pH 7.4 with a half-life of 24 days, provided that enzymic processes do not catalyze lactonization or hydrolysis). Therefore, the rate of transfer of the drug from the stomach to the intestine may not only determine the overall rate of absorption but may also determine the ratio of the acid and the lactone absorbed. These predictions are based on the assumption that the drug transforms with the same rate in the body fluids as in aqueous solution. A recent study of plasma levels of potassium canrenoate in man by Karim *et al.* (26) showed that the plasma levels of canrenoic acid and canrenone were about equal 3 hr. after intravenous administration of potassium canrenoate. This finding indicates that here are enzymatic processes and, hence, an *in vivo* equilibrium is probably established much faster.

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Preformulation Studies of Spironolactone: Effect of pH, Two Buffer Species, Ionic Strength, and Temperature on Stability

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Abstract □ Using a stability-indicating HPLC assay method, the effect of pH, two buffer species (citrate and phosphate), ionic strength, and temperature on the stability of spironolactone in 20% solution of ethyl alcohol in water has been studied. The optimum pH of stability appears to be ~4.5. On increasing the buffer concentration, both species hastened the decomposition of spironolactone. The ionic strength did not affect the stability of the drug. The energy of activation has been estimated to be ~78.8 kJ/mol at pH 4.3. The un-ionized spironolactone is subject to general acid-base catalysis. The K_a and K_{a2} values at 40 °C have been estimated to be 1.63 and 2.8×10^{-5} day⁻¹, respectively. The HPO_4^{2-} ion had ~10 times more catalytic effect than the H_2PO_4^- ion. This data will be used to develop a stable oral liquid dosage form of the drug.

Spironolactone is extensively used in medicine as a diuretic. It is often prescribed to small children, but it is difficult for small children to swallow tablets, the only dosage form available for oral use. A number of methods¹⁻⁴ have been reported for compounding oral liquid dosage forms from the tablets. No systematic study has been reported to determine the physicochemical properties of spironolactone, and to compound a stable oral liquid dosage form of spironolactone using the powder rather than the commercially available tablets.

The purpose of this investigation was to study the effect of pH, phosphate and citrate buffers, ionic strength, and temperature on the stability of spironolactone using a stability-indicating HPLC assay method. These preformulation studies were conducted to help develop an oral liquid dosage form of spironolactone which is not currently available.

Experimental Section

Chemicals and Reagents—All the chemicals and reagents were either USP-NF or ACS grade and used without further purification. Spironolactone powder was generously supplied by G. D. Searle & Company, Skokie, IL, and used as received.

High-Performance Liquid Chromatography—A Waters ALC 202 HPLC system (Waters Associates, Milford, MA) equipped with a multiple wavelength detector (Schoeffel's SF 770) and a recorder (Omnicribe 5213-12, Houston Instruments, Austin, TX) was used. A μ Bondapak phenyl column (30 cm $\times$ 3.9 mm i.d.) was the stationary phase. The mobile phase contained 39% (v/v) of acetonitrile in 0.01 M KH_2PO_4 aqueous buffer. The flow rate was 2.2 mL/min, the detector was set at 254 nm (0.04 AUFS), the chart speed was 80.5 cm/h, and the temperature was ambient.

Preparation of Stock and Standard Solutions—The stock solutions of spironolactone (1.25 mg/mL) and the internal standard, methyltestosterone (0.25 mg/mL) in methanol were prepared fresh every week. These solutions were diluted further with 25% (v/v) aqueous methanol containing 0.01 M KH_2PO_4 as needed. The most commonly used solution (the standard solution) contained 80 $\mu\text{g/mL}$ of spironolactone and 25 $\mu\text{g/mL}$ of methyltestosterone (internal standard).

Assay Solutions—All the assay solutions were diluted with an aqueous buffer solution containing 0.01 M KH_2PO_4 and 25% methanol

(v/v) so that the final concentration of spironolactone was 80 $\mu\text{g/mL}$ (based on the label claim). The concentration of the internal standard in each solution was 25 $\mu\text{g/mL}$.

Preparation of Solutions for Stability Studies—All the solutions for stability studies were prepared by diluting a stock solution of spironolactone in ethyl alcohol (1.25 mg/mL) with an aqueous buffer solution. The final solutions contained 0.25 mg/mL of spironolactone and 20% (v/v) ethanol. All the solutions prepared are listed in Table I. After the zero-day data (assays, physical appearance, and pH values), the solutions were stored at appropriate temperatures (40, 50, and 60 °C) in amber-colored glass bottles (4-oz dispensing bottles, Brockway Glass Company, Brockway, PA). The data were recorded again at the appropriate intervals (between 2 to 156 days).

Results and Discussion

Assay Method—The HPLC assay method is stability indicating, since canrenone (the product of decomposition of spironolactone) eluted before spironolactone. The method is accurate and precise with a percent relative standard deviation of 0.9 based on five readings. The complete details of the developed method are being reported separately.⁶

Effect of pH on the Stability of Spironolactone—The decomposition of spironolactone at all pH values followed first-order law (Figure 1). The pH-rate profile curve (Figure 2) indicates that the optimum pH value of stability for spironolactone is ~4.5. For Figures 1 and 2, the data of solutions 1-7 (Table I) at 40 °C were used. Solutions 8 and 9

Table I—Solutions of Spironolactone (0.25 mg/mL) Prepared in 20% Aqueous Ethanol for Stability Studies

Solution No.	pH ($\pm$ 0.05)	Buffering Agent (M)	Ionic Strength*
1	2.3	Phosphate (0.05)	0.20
2	3.4	Phosphate (0.05)	0.20
3	4.5	Phosphate (0.05)	0.20
4	5.2	Phosphate (0.05)	0.20
5	6.2	Phosphate (0.05)	0.20
6	7.3	Phosphate (0.05)	0.20
7	8.3	Phosphate (0.05)	0.20
8	9.2	Phosphate (0.05)	0.20
9	9.7	Phosphate (0.05)	0.20
10	4.3	Phosphate: citrate ^b (0.14)	0.29
11	4.3	Phosphate: citrate ^b (0.14)	0.38
12	4.3	Phosphate: citrate ^b (0.14)	0.63
13	4.3	Phosphate: citrate ^b (0.14)	0.88
14	4.3	Phosphate: citrate ^b (0.14)	1.13
15	5.3	Phosphate: citrate ^b (0.14)	0.67
16	4.3	Citrate (0.05)	0.30
17	4.3	Citrate (0.10)	0.30
18	4.3	Citrate (0.20)	0.30
19	4.3	Phosphate (0.05)	0.30
20	4.3	Phosphate (0.10)	0.30
21	4.3	Phosphate (0.20)	0.30

* Adjusted with KCl based on the pH of the buffer solution. ^b Prepared according to the directions in ref 6.

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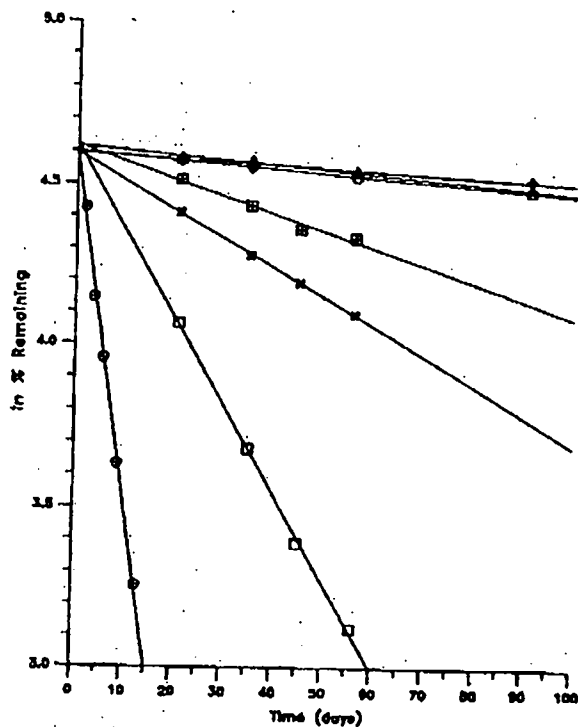


Figure 1—Effect of pH on the stability of spironolactone at 40 °C (solutions 1–7, Table I). Key: (X) pH 2.3; (Δ) pH 3.4; (+) pH 4.5; (O) pH 5.2; (⊖) pH 6.2; (□) pH 7.3; (⊗) pH 8.3.

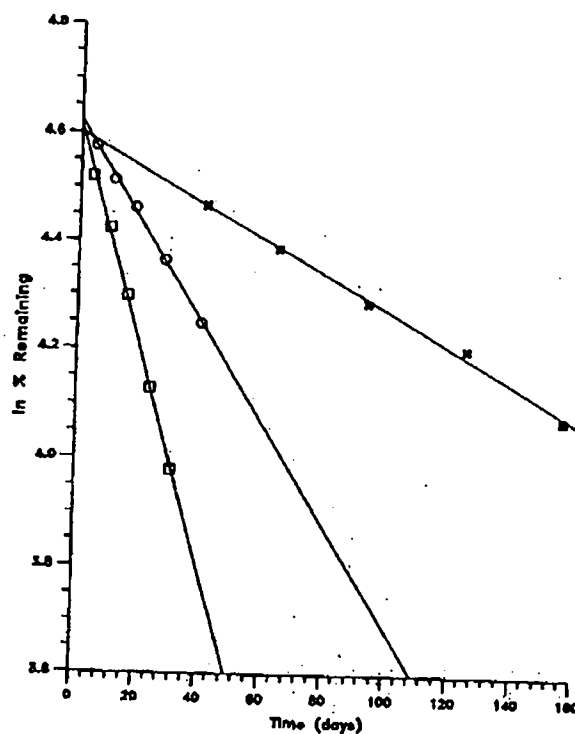


Figure 3—First-order plots of spironolactone at higher temperatures (solution 12, Table I). Key: (X) 40 °C; (O) 50 °C; (□) 60 °C.

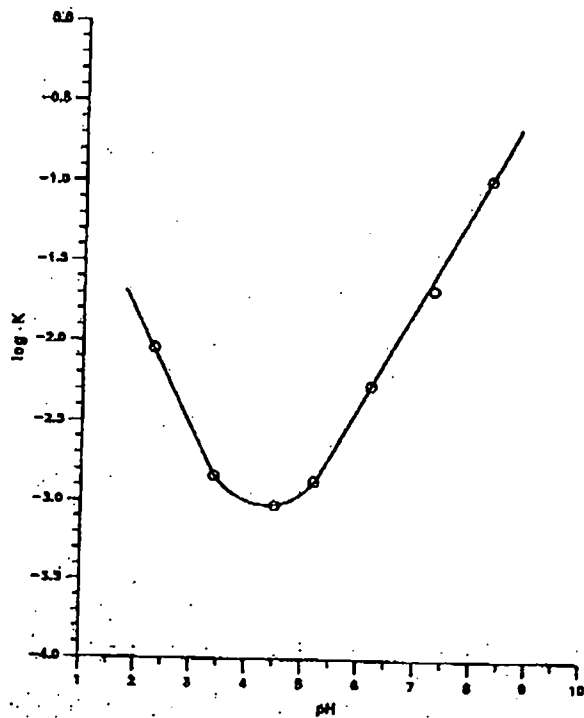


Figure 2—pH-rate profile curve of spironolactone at 40 °C (solutions 1–7, Table I).

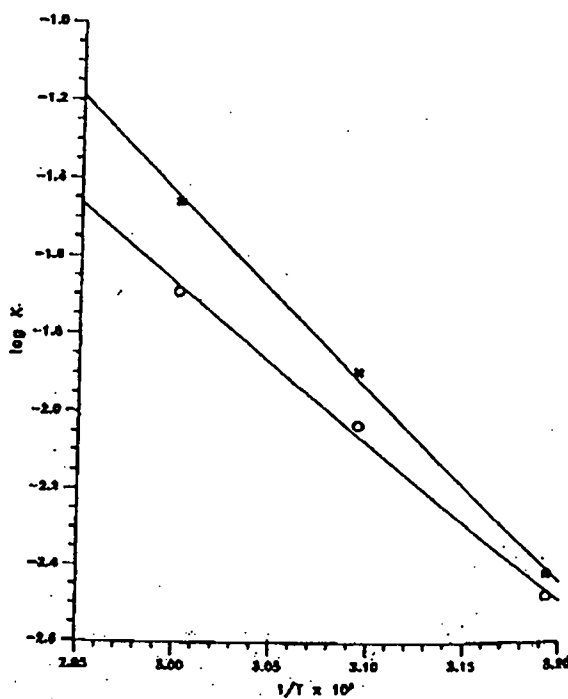


Figure 4—Arrhenius plots for spironolactone at (O) pH 4.3 and (X) 5.3 (solutions 12 and 15, Table I).

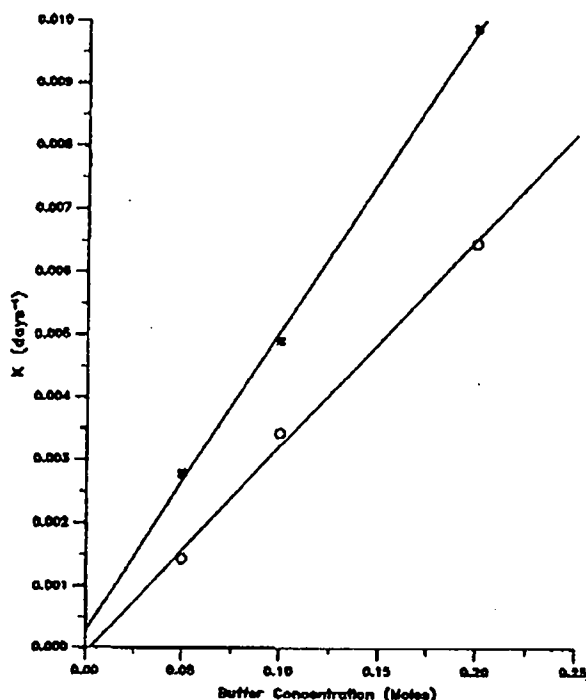


Figure 5—Effect of buffer species on the degradation constant (K) of spironolactone at pH 4.3 and 50 °C (solutions 16–21, Table I). Key: (X) citrate; (O) phosphate.

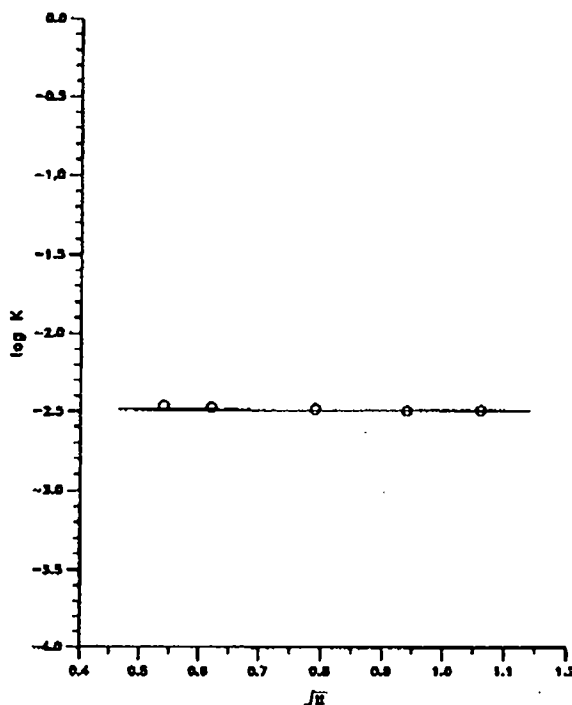
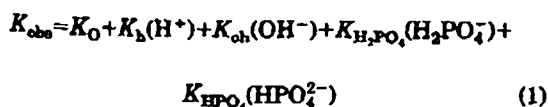


Figure 6—Effect of ionic strength on the stability of spironolactone at pH 4.3 and 40 °C (solutions 10–14, Table I).

(Table I) decomposed rapidly; therefore, they were not studied further.

Effect of Temperature—At all the higher temperatures (40, 50, and 60 °C), the decomposition followed first-order law (Figure 3) and, from the Arrhenius plot (Figure 4), the energies of activation were estimated to be 78.8 and 96.0 kJ/mol at pH 4.8 and 5.3, respectively.

Effect of Buffer Concentrations, Spironolactone Concentration, and Ionic Strength—Both phosphate and citrate buffers hastened the decomposition (Figure 5) of spironolactone with increase in the buffer concentration. Phosphate proved to be only slightly better than the citrate buffer. The slope of the phosphate curve was 0.0333 versus 0.0477 for the citrate curve. The ionic strength did not affect the decomposition constant (Figure 6). The first-order decomposition did not depend on the initial concentration of spironolactone. Since the ionic strength did not affect the K value of spironolactone, it can be assumed that the un-ionized spironolactone reacted with H^+ or OH^- ions. Therefore, the hydrolysis of spironolactone in phosphate buffer may be represented as follows:



Preliminary investigations indicated that the phosphate buffer effect was predominantly due to HPO_4^{2-} . At pH 2.3, the effect of phosphate buffer and the OH^- ion may be neglected

since the concentrations of both these ions at pH 2.3 will be very small. Therefore, eq 1 can be rearranged as follows:

$$K_{obs} = K_O + K_A(H^+) \quad (2)$$

At pH 4.5, the effect of pH and the buffer is at its minimum (Figure 2), therefore, the decomposition is predominantly catalyzed by the solvent. Therefore, the K_{obs} values at pH 4.5 (0.00095 day⁻¹) is K_O . Using the K_{obs} value at pH 2.3 (40 °C) of 0.0091 day⁻¹, and substituting 0.00095 day⁻¹ for K_O in eq 2, the K_A value was estimated to be 1.63 day⁻¹ at 40 °C. On modifying eq 2 by adding $K_{OH}(OH^-)$ and again neglecting the effect of HPO_4^{2-} (since it will be only 1/11 of the total buffer concentration at pH 6.2), the K_{obs} at 40 °C was estimated to be 2.8×10^5 day⁻¹ when a K_{obs} value of 0.00542 day⁻¹ at pH 6.2 was used. The hydrolysis of spironolactone was predominantly subject to OH^- since the value of K_A is $\sim 1.72 \times 10^6$ times more than that of K_A . These preformulation studies will help to develop a stable (for at least 18 months) oral liquid dosage form of spironolactone.

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SHORT COMMUNICATION

INCREASING ORAL BIOAVAILABILITY OF PROGESTERONE
BY FORMULATION

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SUMMARY

Admixture of cholesterol trimethyl acetate to progesterone in proportions that formed solid solutions resulted in significant increase of oral bio-availability in humans. Studies in rats have indicated that increased effectiveness was achieved by increasing lymph absorption.

INTRODUCTION

Absorption of an orally administered drug (i.e., bioavailability) is limited by dissolution of the compound into the aqueous phase in the intestine. In order to increase effectiveness it is necessary to increase dissolution which is directly proportional to surface area; decreasing particle size by micronization was advocated to achieve this [1, 2]. Increased surface area can also result by dispersing low water solubility drugs in water soluble carriers, such as urea [3]. Other investigators described coating of acid-labile drugs (penicillin) with lipids to increase oral bioavailability [4, 5]. Rudel[6] proposed the utilization of sterol lipids to form eutectic mixtures and hypothesized that "the solid solutions...are capable of being absorbed directly by the lymphatic capillary system..." We have investigated intestinal absorption of progesterone and obtained some evidence to support this hypothesis.

EXPERIMENTAL

Sample preparation. 3.75 g of progesterone and 1.607 g of cholesterol pivalate (trimethyl acetate) was deposited by a solvent method (chloroform) on 5.357 g of lactose. Fifty mg of progesterone equivalent was dispensed in hard gelatine capsules (two used per volunteer). For animal studies 200 mg [$4\text{-}^{14}\text{C}$]-progesterone S.A. 480 d.p.m./ μg) was dispersed as microcrystalline suspension in 0.5 ml aqueous carboxymethyl cellulose.

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† Assay sensitivity, 0.1 ng; least detectable amount 0.5 ng.

Human studies. Six healthy male volunteers were used. Bioavailability of both preparations was studied in each; there was a two week interval between each study. Progesterone, 100 mg, was given by mouth in the morning; peripheral blood was collected at 0, $\frac{1}{2}$, 1, 2, 4, 6, 10 and 24 h, plasma was separated and assayed for progesterone using competitive protein binding† (University of Rome).

Animal studies. Rats (Sprague-Dawley strain) anesthetized with ether for the entire procedure were used. Approximately 0.5 cm of the main intestinal lymph duct was exposed by a ventral incision. A single suture was placed under the duct, a silastic tubing (O.D. 1.2 mm, I.D.—0.6 mm, length—15 cm) was inserted and the suture was tied to hold the tubing in place and seal the duct. This technique collects all the lymph draining from the intestines. Samples were then injected into the duodenum and lymph was collected continually to represent 15 min intervals. Peripheral blood was collected every 15 min from a catheter placed in the femoral vein.

RESULTS AND DISCUSSION

Addition of cholesterol pivalate to progesterone increased oral bioavailability in humans. Figure 1 shows progesterone concentration in peripheral plasma (ng/ml, average of six observations) following oral administration of lipid formulated progesterone and lipid-free progesterone. Peak plasma levels of progesterone were seen within 2 h after oral administration; average value at this time was 5 ng/ml of peripheral plasma. Thereafter, the values rapidly declined. After 6 h average value was less than 1 ng/ml; no progesterone was detected at the end of 24 h. When progesterone was given in cholesterol pivalate eutectic

Table 1. Peripheral progesterone plasma levels in male volunteers following oral administration of two progesterone dosages

Time interval, h	2	4	6	10	24
Progesterone on lactose carrier					
Mean, ng/ml (range)*	8.6 (3.8-13.4)	3.2 (1.2-5.2)	1.7 (0.3-31.)	1.5 (0-3.0)	0
(N)†	(8)	(5)	(9)	(10)	(4)
Progesterone: cholesterol pivalate on lactose carrier					
Mean, ng/ml	3.9 (2.5-5.3)	4.9 (3.8-6.0)	3.5 (2.4-4.6)	4.3 (3.5-5.1)	1.9 (1.3-2.5)
(N)	(17)	(16)	(16)	(15)	(12)

* Indicates the lower and upper 95% confidence limits; † (N) number of observations.

Table 2. Absorption of progesterone from the intestinal tract of rats

Dosage form used	15†	30	45	60	75	90
Microcrystalline suspension						
Blood	8.7 ± 2.4	13.8 ± 7.1	18.8 ± 4.2	19.8 ± 3.4	17.8 ± 1.8	18.2 ± 5.6
Lymph	9.7 ± 5.4	18.6 ± 5.3	18.3 ± 5.1	20.7 ± 4.9	19.5 ± 5.0	20.2 ± 5.3
Cholesterol pivalate eutectic						
Blood	4.7 ± 1.9	8.2 ± 3.2	12.4 ± 5.9	14.7 ± 6.0	17.5 ± 4.1	20.6 ± 4.3
Lymph	8.1 ± 4.0	8.2 ± 2.3	13.9 ± 7.5	19.5 ± 4.0	26.6 ± 12.1	32.5 ± 2.4

* Based on total radioactivity measurement. † Time interval, min.

peak plasma levels were seen 4 h after intake (average value 5 ng/ml); the 10-h value was 4 ng/ml, and 2 ng/ml at the end of 24 h, a value significantly different from zero ($P < 0.005$). Estimation of the area under the curve indicated that the bioavailability of progesterone has been increased 4–5 times by admixing the hormone with the sterol. The results are summarized in Table 1.

The results obtained in rats (four animals per group) measuring radioactivity* in lymph and in peripheral plasma during the first 1½ h following the injection of the material into the duodenum are shown in Table 2.

Progesterone appeared rapidly in peripheral blood when microcrystalline suspension was used; the ratio between the radioactivity in lymph and blood was close to unity. In contrast, following injection of progesterone-cholesterol pivalate, eutectic radioactivity levels in the lymph were increased relative to peripheral blood (ratio 1.6).

Several factors limit the effectiveness of orally administered, natural steroid hormones. It is known that their biological half-life is only a few min due to the rapid clearance rate and enzymatic inactivation in the liver. In addition other parameters may decrease intestinal absorption. Nienstedt and Hartiala [7] have shown that progesterone is metabolized in the small intestine and there is considerable enterohepatic recirculation of progesterone metabolites [8]. Our results show that poor oral bioavailability of

natural steroids can be partially overcome by an addition of a cholesterol ester. It is of interest to note that two groups [9, 10] have noted decreased intestinal absorption of sulfadiazine and cortisol in the presence of a hypocholesterolemic bile acid-binding anion exchange polymer (colestipol hydrochloride).

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* [1,2,6,7-³H]progesterone (specific activity 5400 d.p.m./μg) was used. Tissue proteins precipitated with ethanol; aqueous samples were measured in liquid scintillation spectrophotometer.

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Absorption of oral progesterone is influenced by vehicle and particle size

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D4

The oral route of progesterone administration has long been considered impractical because of poor absorption and short biologic half-life. Recent reports suggest that micronization of progesterone enhances absorption and increases serum and tissue levels of progesterone. This study checks serum progesterone levels before and 0.5, 1, 2, 3, 4, and 6 hours after oral administration of 200 mg of progesterone in seven subjects. Progesterone was plain milled, micronized, plain milled in oil, micronized in oil, or micronized in enteric-coated capsules. All patients exhibited a significant increase in serum progesterone levels after oral progesterone administration. Mean peak progesterone levels (30.3 ± 7.0 ng/ml) ($p < 0.005$) were achieved with micronized progesterone in oil at 2.0 ± 0.3 ($p < 0.05$) hours after administration. Four types of oral progesterone had equivalent mean peak elevations and mean times to peak: plain milled, 9.6 ± 2.5 ng/ml at 4.0 ± 0.5 hours; micronized, 13.2 ± 2.4 ng/ml at 3.2 ± 0.4 hours; plain milled in oil, 11.3 ± 3.0 ng/ml at 4.0 ± 0.5 hours; and micronized in enteric-coated capsules, 11.2 ± 3.0 ng/ml at 4.1 ± 0.7 hours. Contrary to traditional teaching, these data show that significant serum progesterone levels can be achieved by oral administration. Absorption can be significantly improved by the physical characteristics of the progesterone and the vehicle used with oral administration. (*Am J Obstet Gynecol* 1989;161:948-51.)

Key words: Progesterone, intestinal absorption, chylomicron, fatty acids

Progestins are used in a variety of clinical situations, including contraception, adjunctive postmenopausal therapy, and treatment of endometrial hyperplasia, luteal phase inadequacy, and premenstrual syndromes.¹ Although orally active, these compounds do not faithfully reproduce the actions of natural progesterone.² Progestins obviate the endometrial hyperplasia and risk of cancer associated with unopposed estrogen therapy for management of the menopause³; however, concern remains that the cardioprotective effect of estrogens may be negated by the adverse effect on lipids caused by the synthetic progestins.⁴⁻⁷ Oral progesterone has not

shown any adverse effect on the beneficial changes in serum lipoproteins induced by the administration of estrogens.^{8,9}

Recent evidence suggests that maneuvers to improve the intestinal absorption of progesterone substantially increase its bioavailability.¹⁰⁻¹² An effective and safe oral alternative to the currently available routes of progesterone administration (vaginal, rectal, or parenteral) would be more practical and acceptable to patients.

This study was designed to test the influence of particle size and vehicle on the absorption and bioavailability of progesterone, as reflected in serial serum progesterone concentrations after a single oral dose.

Material and methods

Progesterone was obtained as either powdered and plain milled or powdered and micronized. One hundred milligrams of either plain milled or micronized progesterone was placed into a No. 3 plain gelatin capsule with or without an alteration in vehicle.

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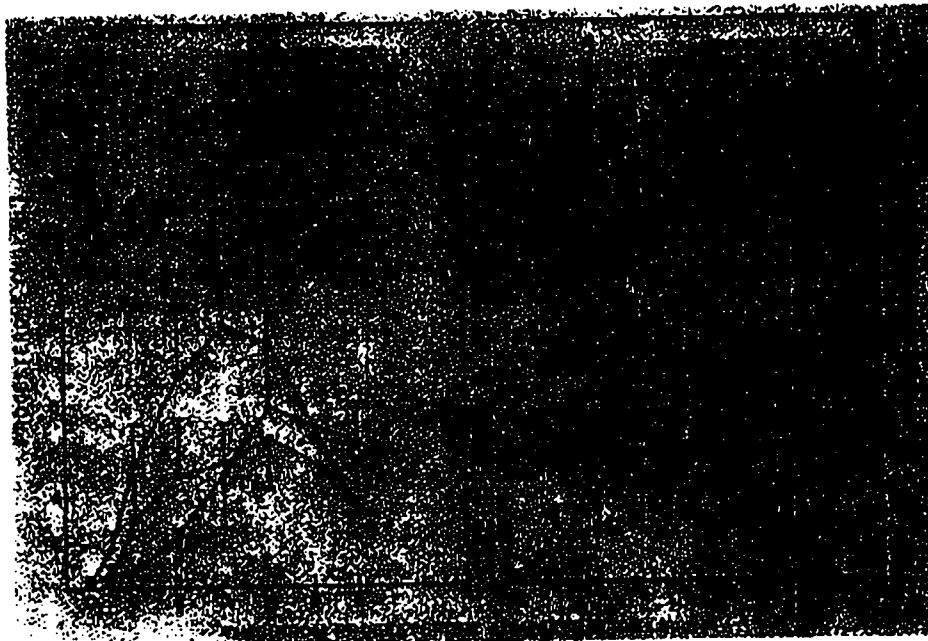


Fig. 1. Serum progesterone concentrations (mean $\pm$ SEM) after oral administration of various preparations of progesterone ($N = 7$).

Five different preparations were made: (1) plain-milled progesterone alone, (2) micronized progesterone, (3) plain-milled progesterone in oil, (4) micronized progesterone in oil, and (5) micronized progesterone in enteric-coated capsules.

Plain-milled and micronized progesterone in oil were prepared by suspending particles of progesterone in a mixture of natural oils that were high in long-chain unsaturated fatty acids. These were placed into No. 8 gelatin capsules for a total steroid concentration of 100 mg per capsule.

Enteric-coated capsules were prepared by inserting 100 mg of powdered micronized progesterone into similar capsules and then covering the capsule with a waxy coating of methylcellulose, carnauba wax, and stearic acid.

Six postmenopausal women and one man volunteered for oral administration of each of these preparations. Each subject was tested six times, twice with micronized progesterone and once each with micronized progesterone in oil, micronized progesterone in enteric-coated capsules, plain-milled progesterone alone, and plain-milled progesterone in oil. At least 3 days was required between each absorption study. For each absorption study, each individual had fasted overnight and was given a 200 mg dose (two capsules) orally of the study progesterone preparation. The fast was continued for 1 hour after ingestion of progesterone, then regular food and fluid intake was allowed. Only sedentary activity was permitted during the study pe-

riod. Phlebotomy was performed through a heparin lock at baseline, 0.5, 1, 2, 3, 4, and 6 hours after capsule administration.

Each serum sample was placed on ice and then serum was separated by centrifugation, labeled, and frozen for determination of progesterone. The assays for progesterone were performed by radioimmunoassay with use of commercial kits obtained from Diagnostic Products Corporation, Los Angeles, Calif. Intraassay variation for progesterone was 7.5%, and interassay variation was 4.3%. Normal midluteal progesterone concentrations for this radioimmunoassay had been previously determined."

Data were analyzed statically with Fisher's least significant difference test, Scheffé's multiple comparisons, and the paired t test. The areas under the curves were generated with a modification of the CLINFO computer program at Vanderbilt University Medical Center.

Results

All preparations administered orally resulted in a significant absorption of progesterone as detected by radioimmunoassay for progesterone in serum. Mean serum levels of progesterone at each time point are summarized in Fig. 1.

Micronized progesterone in oil showed the highest average progesterone concentration (30.3 ± 7.0 ng/ml, mean $\pm$ SEM) and the shortest time from ingestion to measured peak (2.0 ± 0.9 hours, mean $\pm$

Table 1. Absorption of progesterone according to particle size and vehicle

Progesterone	n	Peak progesterone Conc (ng/ml) (mean $\pm$ SEM)	Time to peak (hr) (mean $\pm$ SEM)
Micronized progesterone	14	13.2 $\pm$ 2.4	3.2 $\pm$ 0.4
Micronized progesterone in oil	7	30.3 $\pm$ 7.0*	2.0 $\pm$ 0.3†
Micronized enteric-coated progesterone	7	11.2 $\pm$ 3.0	4.1 $\pm$ 0.7
Plain-milled progesterone	7	9.6 $\pm$ 2.5	4.0 $\pm$ 0.5
Plain-milled progesterone in oil	7	11.3 $\pm$ 3.0	4.1 $\pm$ 0.5

* $p < 0.005$.† $p < 0.05$.

SEM) (Table 1). No significant difference was found for either peak concentration, time to peak, or mean area under the curve between the other preparations.

Inpatient variability was examined by the repetition of an absorption study with micronized progesterone (Fig. 1, A). Although the peaks were higher in six of seven patients in the initial study compared with the second series (16.98 ± 3.5 vs. 9.5 ± 2.9 mg/ml, mean $\pm$ SEM), no statistically significant difference was identified. The time to peak concentration was faster, but not significantly different, during the first series (2.5 ± 0.4 vs. 4.0 ± 1.7 hours, mean $\pm$ SEM).

By the end of the study (6 hours after administration of progesterone), all groups demonstrated persistently increased serum progesterone concentrations as compared with baseline levels. Despite a significant difference in peak level and area under the curve for micronized progesterone in oil, serum progesterone concentrations at 6 hours were equivalent in all preparations (Fig. 1, B). No untoward effects to the progesterone preparations were experienced by any of the study subjects.

Comment

The oral route of progesterone administration has long been considered clinically ineffective, because of reported poor gastrointestinal absorption and short biologic half-life. However, Kindl et al.¹³ demonstrated that a preparation consisting of nonmicronized progesterone on a lactose carrier in gelatin capsules rapidly increased serum progesterone concentrations in male volunteers. With this formulation, absorption was rapid and reached low luteal phase levels with a mean of 8.6 ng/ml (range 3.0 to 13.4 ng/ml) at 2 hours and a rapid decline to 3.2 ng/ml by 4 hours.

Later, Whitehead et al.¹⁰ described oral administration of nonmicronized progesterone to five postmenopausal women. After daily administration for five consecutive days, peak progesterone concentrations of 9.5 ng/ml were attained. In these subjects a sustained elevation of progesterone was maintained for more than 36 hours after the 5-day loading dose.

Chemical and physical alterations of the steroid molecule and the vehicle used with oral administration significantly affect absorption. Kindl et al.¹³ described two methods that theoretically increased the bioavailability of oral progesterone in rats. First, progesterone concentrations were increased through techniques that enhanced progesterone absorption through the intestinal lymphatics. Second, these investigators showed that a decrease in the particle size of progesterone through micronization increased aqueous dissolution in the intestine and further enhanced absorption.

Lipophilic molecules are absorbed from the intestinal tract and transported to the general circulation by a variety of mechanisms. Most lipophilic molecules that are transported through the lymphatics reside in the triglyceride core of the chylomicron. The properties of the vehicle in which a drug is administered have been described to affect the degree of lymphatic absorption. With oils, the greater the degree of unsaturation of the fatty acid, the more rapid the onset of chylomicron synthesis. Cheema et al.¹⁴ found that linoleic acid and arachis oil produced the highest concentration of chylomicron in the lymph in rats. Avoidance of the extensive prehepatic clearance of progesterone through absorption into the portal circulation should thereby enhance bioavailability of oral progesterone.

Numerous clinical studies of serum progesterone levels after oral administration of micronized progesterone are now available.^{10-12, 15} Progesterone taken orally is physiologically active, producing a significant increase in tissue progesterone concentrations in breast, endometrium, and myometrium.¹⁶ Orally administered progesterone also induces histologic and biochemical changes in the endometrium after 10 days of administration in estrogen-primed postmenopausal women.¹⁷ Concomitant oral administration of estradiol and progesterone for 1 year did not cause abnormal proliferation or hyperplasia of the endometrium.¹⁸

The current study evaluated the combination of the processes of micronization and suspension in long-chain fatty acids as means of influencing the absorption of progesterone administered orally. These data clearly indicate a synergistic effect of these two processes. Of particular interest was the observation that the suspension of nonmicronized progesterone in oil did not significantly enhance circulating progesterone concentra-

tions over either micronized or plain-milled progesterone. In addition, the process of enteric coating to protect against gastric acidity did not increase absorption of progesterone. Although sedative and hypnotic properties of oral progesterone have been reported, none of the subjects participating in this study experienced these effects.¹⁰

On the basis of the results of this study, the optimal preparation for the administration of natural progesterone should include micronization of the progesterone particles and dissolution in oils consisting of principally long-chain fatty acids.

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Atrial natriuretic peptide during the menstrual cycle

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Plasma atrial natriuretic peptide levels were found to decrease from 82.6 pg/ml in the follicular phase to 55.8 pg/ml in the luteal phase in 10 normal menstruating women. The reduced level of plasma atrial natriuretic peptide could be a compensatory response to the natriuretic effect of the increased plasma progesterone in the luteal phase. (*AM J OBSTET GYNECOL* 1989;161:951-2.)

Key words: Atrial natriuretic peptide, menstrual cycle, progesterone

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Influence of cohesive properties of micronized drug powders on particle size analysis

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Abstract: Particle size analysis results with respect to micronized, mean particle size below 10 μm , furosemide, chloramphenicol palmitate and acetaminophen particles are dealt with in this paper. Special consideration was given to the effect of the agglomeration of particles on data generated by three size measurement techniques. The physico-chemical basis for preparing sufficiently well dispersed and stable suspensions for analysis by employing mechanical methods of pretreatment are shown. Furthermore, methods to determine the state of dispersion and methods to assess the individual particle size before size analysis are described. An attempt was also made to establish the statistical confidence that can be assigned to a particular instrument and the confidence level that may be placed on comparative data obtained with the different particle size analysers. Results especially showed the impact of the agglomeration of very small furosemide particles, mean size 3 μm , on particle size analysis and the importance of controlling the cohesive properties of this drug. To overcome the problems associated with agglomeration more attention must be paid to the physical properties of the drug substance. Combining particle size analysis with bulk density, surface area and microscopical studies also helped to identify potential problems.

Keywords: Particle size analysis; drugs; cohesive powders; micronized powders.

Introduction

The importance of particle size to the pharmaceutical industry is widely recognized and this is reflected in that particle size analysis is a commonplace activity [1]. The size distribution of drug powders is a key property that can influence its transport, storage, reactivity and solubility [2]. In pharmacy we are mostly concerned with particles, ranging from 0.1 to 1000 μm in size, that only approximate to spheres in shape. The degree of fineness of a powder is of outmost importance since the presence of very small particles can modify the cohesive properties of the powder [3]. In practice, the primary particles of micronized powders, — particle size below 20 μm — will adhere to one another due to surface forces forming secondary particles, agglomerates and aggregates [4].

There are close to 200 different methods for characterizing some size-dependent aspects of a fine particle system [5]. Recently, laser diffraction has become one of the most widely used methods for determining particle size [2]. Its popularity arises out of the apparent simplicity and because a size distribution may be obtained within minutes. Selection of a specific

size analysis method may be limited by pharmacopoeia requirements, but in general the most efficient method should be selected on particle properties, for example cohesiveness, wettability and solubility [6]. Recently the tendency is to extract information on the effect of a specific particle property from the comparison of data obtained with different particle sizing techniques [7].

An important precondition for most methods of particle size analysis is the preparation of suspensions with a defined state of dispersion [8]. The preparation of a suspension of deagglomerated particles is particularly difficult when working with micronized, cohesive powders. Dispersion is intended to counteract the forces between particles by physico-chemical and mechanical means such that the particles finally exist individually. To achieve this the following three fundamental processes must take place: (1) wetting of the solid; (2) deagglomeration of particle agglomerates; and (3) stabilization of the dispersed suspension [4]. Some mechanical means used to disperse particles include, stirring, shaking and ultrasonic treatment of the suspension [8]. Most commercially available particle size analysers employ one or the other sample

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suspension and dispersion technique during analysis.

This investigation attempted to study the influence of the cohesive behaviour of micro-fine drug particles on particle size analysis. It was done by comparing the differences between particle size analysis of micronized furosemide, chloramphenicol palmitate and acetaminophen particles in the same and in different instruments. The aim was also to evaluate methods used to assess agglomeration and methods employed to disperse agglomerates before analysis.

Materials and Methods

Materials

Three micronized drugs, furosemide, chloramphenicol palmitate and acetaminophen were chosen due to differences in cohesive behaviour. The powders were supplied by Fine Chemicals (South Africa) and complied to USP standards. Larger particles (mean size approximately 20 μm) were prepared by milling large particles, recrystallized from hot saturated solutions (50°C) of the drugs in ethanol, with a Retz high speed mill (Haan, Germany). X-ray powder diffraction analysis showed that the crystal forms of the recrystallized material were similar to that of the supplied powder. Powders were stored in dessicators over silica gel. All the solvents and chemicals were analytical grade and water fit for liquid chromatography was used.

Particle size analysis methods

Three different particle size techniques were selected:

- (1) the electrical sensing zone technique (Coulter Counter, model ZB, England) which measured an equivalent volume diameter of particles suspended in an electrolyte solution and held in suspension by a propeller stirrer;
- (2) the laser diffraction spectrometry method (Sympatec, Germany) whose output is a volume (or mass) distribution. Powders were suspended either with the aid of a low frequency ultrasonic bath or a dry powder dispersing attachment. This attachment permits the application of laser diffraction to study dry powders with particle sizes from less than 0.5 μm ;
- (3) the dual discipline analysis technique (Galai-Cis-1, Israel), integrating diffraction

and image analysis, for particle sizing whose output is also a volume (or mass) distribution. Suspended particles were measured in a small glass cuvette. A small magnetic stirrer inside the cuvette prevented sedimentation of the particles during analysis.

The criteria for the selection were to apply modern equipment commonly used for routine analysis. The operating principles of these instruments have been described in papers and textbooks and will be omitted here. It should be pointed out that they all perform fast analyses, originate highly reproducible results and are capable of handling a large variety of materials. The dry powder method, however, uses large quantities of powder.

Validation of particle size analysis

The particle size analysers were calibrated for particles in liquid analysis using polystyrene spheres of known diameter, 2 and 20 μm . Measured sizes obtained with the three instruments were not significantly different from data supplied by the manufacturer of the standard particles, with values of $2 \pm 0.3 \mu\text{m}$ and $20 \pm 2.4 \mu\text{m}$, respectively. As the dry dispersion apparatus, coupled to the Sympatec particle size analyser, requires a large volume of powder for analysis it was not possible to validate the instrument with the standards. Furthermore, to check on the dissolution of the smaller drug particles relative to the larger particles, the number of particles were counted before and 5 min after each measurement. Comparison of these counts indicated that sample dissolution was negligible.

Methods used to disperse powders

Dispersion into air was performed with a dry powder disperser, attached to the Sympatec Helos particle size analyser, using compressed air. When using the particles in liquid method, concentrated homogeneous suspensions were prepared either in an ultrasonic bath or by lightly shaking suspensions with a mechanical shaker. Suitable non-solvent dispersing liquids were chosen as a result of preliminary tests. Both membrane filtered solutions saturated with the drug and saturated solutions containing 0.011 g l^{-1} polyoxyethylene sorbitan monooleate were used as dispersing solutions [9]. For particle size analysis with the Coulter Counter a 1% calcium chloride electrolyte solution was saturated with the drug.

Microscopy

Microscopy and especially electron microscopy is the best method for determining the particle size directly because it gives additional information on particle shape and aggregates [6]. The powders were studied with a Cambridge Stereoscan 250 scanning electron microscope (SEM). Electronmicrographs and video images, taken with cameras attached to the electron microscope, were studied and the particle sizes assessed.

Surface area measurement

The surface area (A) of a powder constituted by spheres is universally related to the size of the individual particles by the following equation [10]

$$d = 6/(\rho A), \quad (1)$$

where ρ is the density of the solid and d is the mean diameter. For actual powders comprised of particles of different sizes and irregular shapes the relationship is of course more complicated but the equation still provides a rough guide to the order of magnitude of particle size [11]. The most widely applied method for surface area measurement is the BET method. A micromeritics model 2000 (Micromeritics, USA) high speed surface area analyser, designed for single point BET, using fixed pressures and variable volumes was used to measure the surface areas of the micronized powders. Results are the mean of three determinations.

Density measurement

An indirect method to measure the cohesiveness of a powder from bulk density is to calculate the percentage compressibility (C) used to evaluate the flow properties of powders [12].

$$C = (D_t - D_p)/D_t \times 100, \quad (2)$$

where D_t is the tapped density and D_p the poured or fluffed density. Powders with a compressibility of $>28\%$ have extremely poor flow properties and are classified as cohesive [13]. To measure the density change a known powder mass was placed in a calibrated glass cylinder and the cylinder mechanically tapped until no change in the volume could be detected. The true density was measured using an air comparison pycnometer (Model 930,

Beckman, USA). Presented values are the mean of five determinations.

Calculations and statistical interpretations

For fine particulate pharmaceutical powders, the volume or mass bias (identical to volume if the measured particles have equal densities) proved the most useful way of representing particle diameters of milled or micronized powders [6]. These powders usually have log-normal particle distributions that can be described completely by the geometric median or mean diameter plus geometric standard deviation. To define the size distributions and compare the characteristics of the powders measured size distributions were broken down into different size ranges and the data converted to the volume (or mass) distribution [6]. Results presented throughout are the mean of five individual measurements and particle size distributions are the relative frequency distributions by volume [6]. Mean volume particle sizes were compared according to the Newman-Keuls test (CSS: Statistica, Statsoft). A 95% confidence level ($P \leq 0.05$) was considered satisfactory for indicating significant differences in mean volume particle size.

Contact angle measurement

To lower the interfacial tension surfactants (wetting agents) are used [14]. The most popular approach to obtain an indirect contact angle for powders is to prepare a compressed disc and to observe a small drop of liquid on the surface. Powder discs were compressed on a RIIC ring press used for compressing discs for infrared spectroscopy. For each powder three discs were prepared and the contact angle of the dispersing solutions measured. Results are the mean of 15 measurements per disc.

Results and Discussion

No uniform behaviour of the powders could be detected when examining the appearance of the micronized, and recrystallized, milled, furosemide, chloramphenicol palmitate and acetaminophen powders. Estimation of the cohesive properties of the powders, indicated by a percentage compressibility, equation 2, larger than 28%, showed that the micronized furosemide powder was extremely cohesive ($C = 42\%$). Micronized chloramphenicol

palmitate ($C = 30\%$) and acetaminophen ($C = 26\%$) powders were also cohesive. The larger recrystallized, milled powders could all be classified as fair to free flowing powders. $C = 21\%$ for furosemide, $C = 11\%$ for chloramphenicol palmitate and $C = 7\%$ for acetaminophen.

The scanning electronmicrographs of the micronized furosemide powder, Fig. 1, show that the powder contained agglomerates of particles with varying sizes. Such electronmicrographs and video images taken of all the powders, were used to estimate the microscopical mean length of the powders, listed in Table 1. However, microscopical examination was necessarily confined to a minute sample of the material, so that large numbers of such samples had to be examined to obtain meaningful results. Notwithstanding highly accurate

values for particle size not being attainable the particle size assessed from carefully studied micrographs proved an ideal tool for assessing the mean particle size before analysis with a dedicated particle size analysis instrument. Particle sizes were also estimated from surface area measurements, equation 1. When a solid is cohesive, the particle size calculated from the external surface area, may be smaller by several orders of magnitude than the particle size measured without dispersion of agglomerates. Surface area calculated mean sizes for the three micronized powders are listed in Table 1. Despite the arbitrariness of the correlation between surface area and particle size it also indicated primary particle size. For practical reasons the application of the BET method to the study of surface area had to be limited to particles that were extremely finely divided.



Figure 1
Scanning electronmicrograph of agglomerated, micronized furosemide particles with a mean volume particle size of $3\text{ }\mu\text{m}$.

Table 1
Powder and particle properties of the drug powders

Drug powder	Contact angle (°)	Powder density (g cm^{-3})	Surface area ($\text{cm}^2 \text{g}^{-1}$)	Mean size surface area (μm)	Microscopical mean length (μm)
Furosemide	81	1.63	11200	3	5
Chloramphenicol palmitate	122	1.25	6300	3	8
Acetaminophen	69	1.25	7600	6	6

Taking into account the cohesive behaviour of the micronized powders, it was, therefore, essential that these powders were completely dispersed before particle size analysis with the dedicated particle size analysers. To obtain and maintain a high degree of dispersion of the powders in the dispersing liquids the solid had to be wet by the dispersing liquid as spontaneously and completely as possible. Furthermore, the forces of repulsion had to be as high and Van der Waals energies of attraction as low as possible. For the characterization of the state of dispersion a number of methods were used including measurement of wettability from contact angle measurements, microscopical determination and comparison of the difference in particle size measured before and after dispersion.

The contact angles of saturated aqueous solutions on compressed discs of the three drugs, listed in Table 1, showed that the drugs were not easily wet by water but completely wet by saturated solutions containing a surfactant, polyoxyethylene sorbitan monooleate, below the critical micelle concentration. Contact angles could not be measured because the dispersing solutions spread over the disc surfaces. To enhance and increase dispersion, suspended powders were subjected to sonication. Ultrasonic treatment of suspensions is presently the most common and the most effective method used for agglomerate breakdown [8]. The effect of sonication time on the mean volume particle size of the agglomerated, micronized powders, measured with the Galai-

Cis-1 instrument, is illustrated in Fig. 2. The mean volume particle diameter of all the powders decreased with increased sonication time. This decrease in size was especially significant for the agglomerated furosemide powder shown in Fig. 1. When agglomerates of furosemide particles were not fully disintegrated into constituent particles, big loose aggregates were observed in the samples and the particle size distributions were multi-modal with an average size above 100 μm . After 12 min of sonication for the furosemide, 4 min for the chloramphenicol palmitate and one minute for the acetaminophen micronized powders the mean volume particle size and distribution remained constant. These times were considered as the minimum time necessary to ensure the optimum degree of disintegration and dispersion before size analysis. The larger size powders were all dispersed after one minute sonication in the surfactant dispersing solutions. The degree of sample dispersion was confirmed by microscopical evaluation of suspensions.

The results obtained from dispersion evaluation confirm that except for micronized furosemide particles and large acetaminophen particles, sample sonication in a surfactant dispersing solution disperses the agglomerates into single particles available for counting and sizing. Sonication was not sufficient to completely disperse furosemide agglomerates because even after 16 min the particle size distribution was bi-modal with a mean size of 4 μm , compared to the single population with

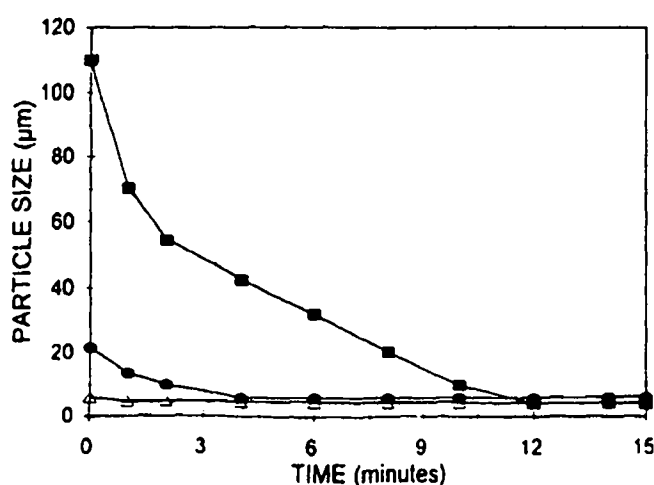


Figure 2
Effect of sonication on the mean volume particle size, measured with the Galai-Cis-1 particle size analyser, of micronized (■) furosemide, (●) chloramphenicol palmitate and (Δ) acetaminophen powders and suspended in saturated aqueous drug solutions containing a surfactant.

a significantly smaller mean volume particle size, 3 μm , measured after dry powder dispersion, Fig. 3. The observed decrease in particle size of the acetaminophen after sonication for longer than 2 min, and because sonication is known to fracture particles, implicated that sonication resulted in primary particle breakdown of particularly larger acetaminophen particles. Dry powder dispersion also led to primary breakdown of the larger sized acetaminophen particles.

Subsequently, the mean volume particle size distribution of each of the six powders was measured without dispersion and after dispersion in surfactant solutions with sonication, using each of the three selected particle size analysers. The particle size of powders measured after being suspended in drug saturated aqueous solutions and dispersed by shaking the suspension for 1 min with a low intensity mechanical shaker were regarded as representing the particle size of the powders when not dispersed. Results are listed in Table 2 for the small sized micronized powders and Table 3 for the larger recrystallized, milled particles. For all the micronized powders the sizes measured without dispersion were significantly larger than after dispersion irrespective of the method of analysis used, Table 2. This was also true for the larger furosemide particles, Table 3.

The particle size of the micronized furosemide powder after dispersion, Table 2, depended on the method used for analysis. The mean size measured with the electrical sensing

zone technique was significantly larger, 12 μm , than obtained with the laser light scattering and light blockage instruments, average 4 μm ($p = 0.046$). The smallest mean volume size, 3 μm , was measured with the laser light scattering instrument after dry dispersion. This size, although smaller, did not differ significantly from the sizes obtained with the particle in liquid methods, average size 3 μm , but differed significantly from that measured with the electrical sensing zone technique, 12 μm . The size distribution obtained after dry dispersion was also the only uni-modal dispersion, Fig. 3, measured for the micronized furosemide powder. In comparison bi-modal and multi-modal distributions, Fig. 3, were obtained with the other methods. According to its compressibility, Table 1, the larger furosemide particles were also cohesive and once again the particle size measured before dispersion was significantly larger than after dispersion.

Micronized chloramphenicol palmitate powder was not as cohesive as the furosemide powder and, therefore, provided to be less sensitive to changes in the particle size analysis methods. After dispersion the particle sizes measured with the different methods were similar. The difference between the size measured before, average $\pm 20 \mu\text{m}$, and after dispersion, average $\pm 6 \mu\text{m}$, is not as significant as for the furosemide powder. The larger particle size chloramphenicol palmitate powder, Table 3, was not cohesive and the size measured after dispersion was not significantly

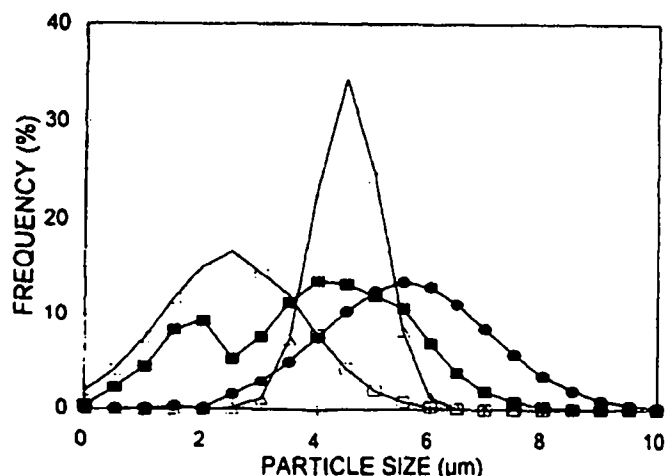


Figure 3
Relative percentage frequency particle size distribution by volume of the micronized (■) furosemide (●) chloramphenicol palmitate and (Δ) acetaminophen powders measured with the laser light scattering instrument and suspended in surfactant solutions plus the distribution of the furosemide powder after dry dispersion (□).

Table 2
Mean volume particle size and standard deviation of micronized drug particles measured before and after dispersion using different particle size analysis techniques. Results are the mean of five measurements

Size analysis method	Furosemide		Chloramphenicol palmitate		Acetaminophen	
	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)
Electric sensing zone*	126 $\pm$ 23.3	12 $\pm$ 4.2	25 $\pm$ 6.3	6 $\pm$ 1.6	8 $\pm$ 2.0	5 $\pm$ 0.5
Light blockage†	110 $\pm$ 26.3	4 $\pm$ 1.5	22 $\pm$ 5.5	6 $\pm$ 1.5	6 $\pm$ 1.5	5 $\pm$ 0.5
Laser light scattering‡	94 $\pm$ 19.0	3 $\pm$ 1.3	19 $\pm$ 5.3	6 $\pm$ 1.5	5 $\pm$ 1.5	5 $\pm$ 1.6
Dry powder analysis§		3 $\pm$ 1.2		5 $\pm$ 0.4		4 $\pm$ 1.4

* Coulter Counter.
† Galai-Cis-1.
‡ Sympatec Helos.
§ Sympatec Helos plus Rodos dry disperser.

Table 3
Mean volume particle size and standard deviation of larger drug particles measured before and after dispersion using different particle size analysis techniques. Results are the mean of five measurements

Size analysis method	Furosemide		Chloramphenicol palmitate		Acetaminophen	
	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)	Not dispersed ($\mu\text{m} \pm \text{SD}$)	Dispersed ($\mu\text{m} \pm \text{SD}$)
Electric sensing zone*	42 $\pm$ 11.5	23 $\pm$ 5.1	22 $\pm$ 6.4	22 $\pm$ 5.9	25 $\pm$ 6.5	19 $\pm$ 5.5
Light blockage†	33 $\pm$ 7.7	21 $\pm$ 4.6	24 $\pm$ 6.0	20 $\pm$ 5.2	26 $\pm$ 6.2	16 $\pm$ 5.2
Laser light scattering‡	30 $\pm$ 10.9	20 $\pm$ 4.4	21 $\pm$ 5.6	20 $\pm$ 5.3	19 $\pm$ 6.1	16 $\pm$ 4.1

* Coulter Counter.
† Galai-Cis-1.
‡ Sympatec Helos.

different from that measured before dispersion. According to differences in particle size measured before and after dispersion, listed in Table 2 and 3, the acetaminophen powders were apparently not cohesive. However, microscopical evaluation, did show that a slight decrease in particle size after sonication and dry powder dispersion could be the result of primary particle fracture. The breakdown of primary particles by sonication was especially significant for the larger acetaminophen particles. Table 3.

Throughout the particle size distributions obtained with different size analysers showed dispersion of cohesive particles before analysis affected the measured mean size and distribution. The mean particle size and distribution of the cohesive, poorly wettable powder, furosemide, in particular varied considerably depending on the method used for particle size analysis, especially the method used to disperse agglomerates before measurements. This variation was not as significant for less cohesive chloramphenicol palmitate powder and was almost non existent for the free flowing acetaminophen particles.

Conclusions

This investigation attempted to study the influence of the cohesive behaviour of micro-fine particles on particle size analysis. It was done by comparing the differences between analysis of different materials in the same instrument and the same material in different instruments. Results illustrated the sensitivities of the different sizing techniques to the presence of particle agglomerates or flocculates and demonstrated the effects of improper dispersion on particle size distribution results. It showed that more attention should be paid to physical properties of the drug substance and that the combination of particle size analysis with bulk density, surface area and scanning electron microscopy studies helped to identify potential problems. To be able to determine the particle size distribution of a cohesive powder the particle size and distribution of the individual particles had to be known because when a powder with fine particles of unknown size was considered, then many distributions composed of both separated particles and/or agglomerates with different sizes could not be identified.

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Throughout the study particle size control proved only truly effective when the following was achieved: an easily standardized analysis method and dispersion technique was used; the method was applicable to primary particle size assessment and sensible interpretation, presentation and comparison of particle size data were possible. For cohesive powders the question also arises: which dispersing state of the system must be reached with respect to the measuring aim? The fulfilment of the following requirement proved to be ideal: the solid particles should be contained in the suspension as individual particles, the dispersing state must not change during particle size analysis and the individual particle properties must not be altered during dispersion or analysis.

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D12

United States Patent [19]
Ester

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[45] **Date of Patent:** May 22, 1990

- [54] **FORMULAE AND METHODS FOR
SUBLINGUAL INGESTION OF NATURAL
PROGESTERONE**
- [76] **Inventor:** George C. Ester, 9457 118th La., N.
Seminole, Fla. 33542
- [21] **Appl. No.:** 87,320
- [22] **Filed:** Aug. 20, 1987
- [51] **Int. Cl.³** A61K 31/56
- [52] **U.S. Cl.** 514/177; 514/899;
514/951; 514/960
- [58] **Field of Search** 514/951, 960, 899, 177
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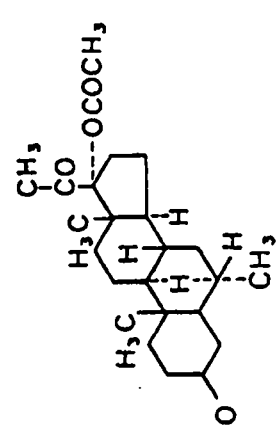
Primary Examiner—H. M. S. Saeed
Assistant Examiner—J. Saba

[57] **ABSTRACT**

Formulae, means, and methods for the sublingual ingestion of natural Progesterone comprising milling and blending natural Progesterone such that it can be absorbed by the sublingual mucosae of a patient and applying it to said sublingual mucosae in a form which permits such absorption.

12 Claims, 5 Drawing Sheets

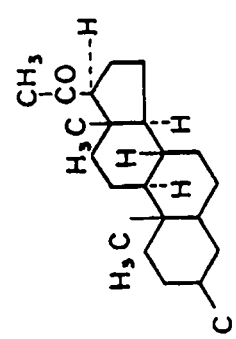
375.



17-Hydroxy-6 α -methylpregn-4-ene-3,20-dione acetate [71-58-9] C₂₄H₃₄O₄ (386.53)

FIG-1

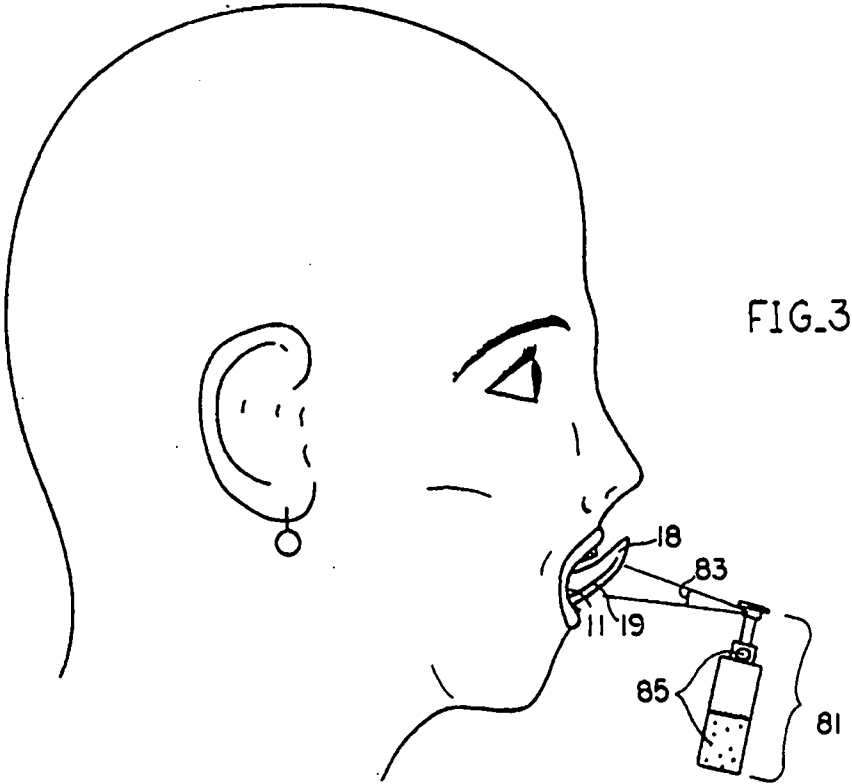
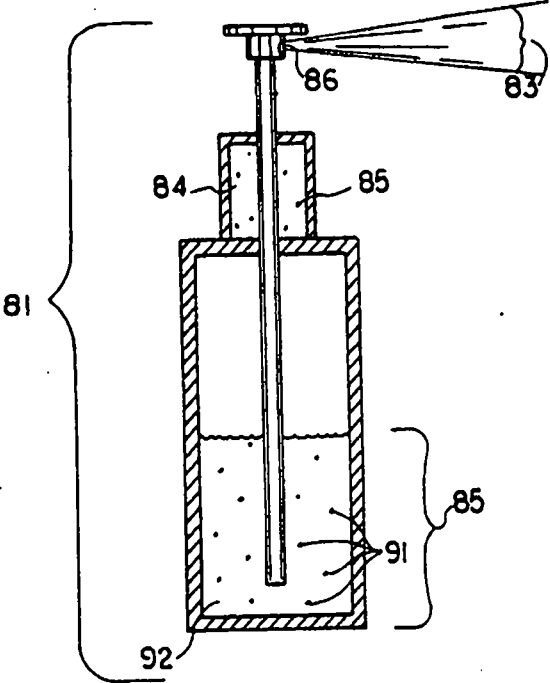
Pregn-4-ene-3,20-dione;
Progesterone USP



Progesterone [57-83-0] C₂₁H₃₀O₂ (314.47)

FIG-2

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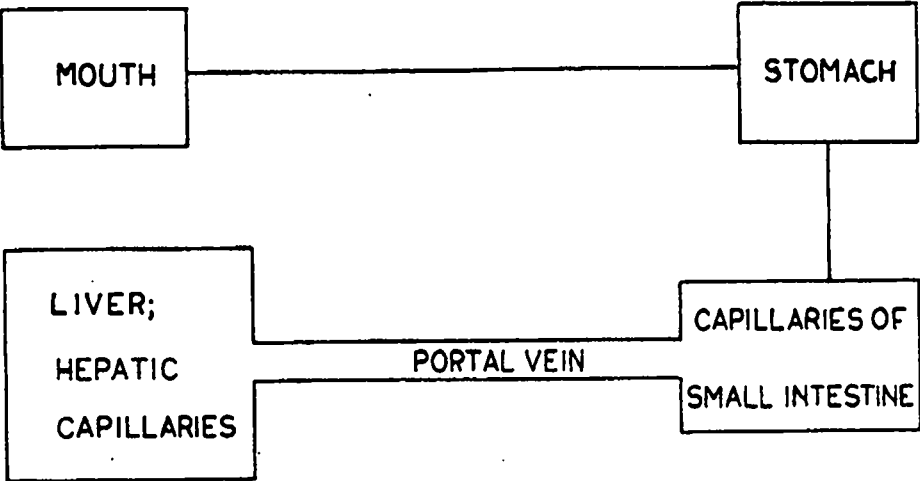


FIG_4

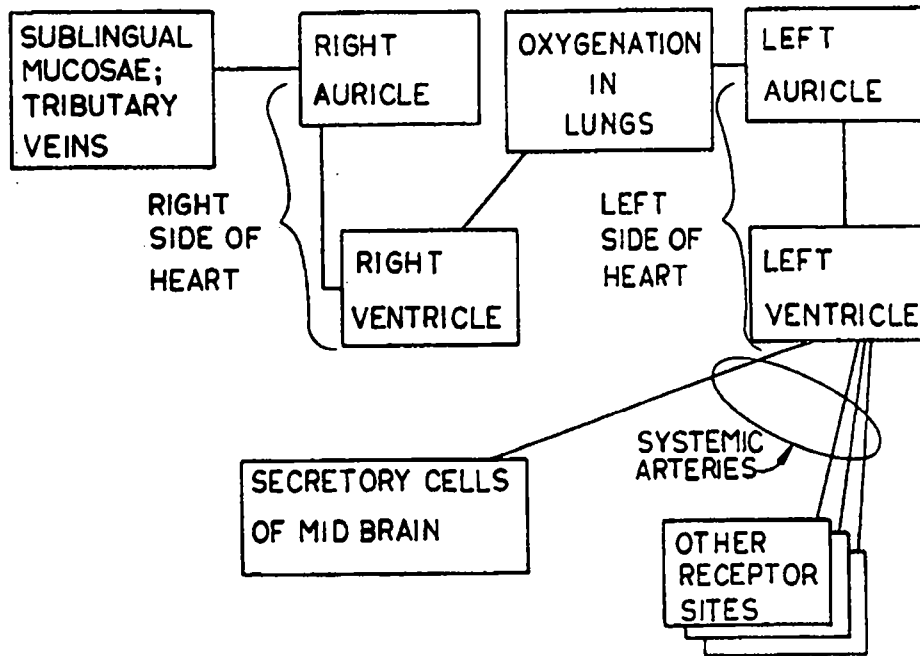


FIG_5

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FIG. 6

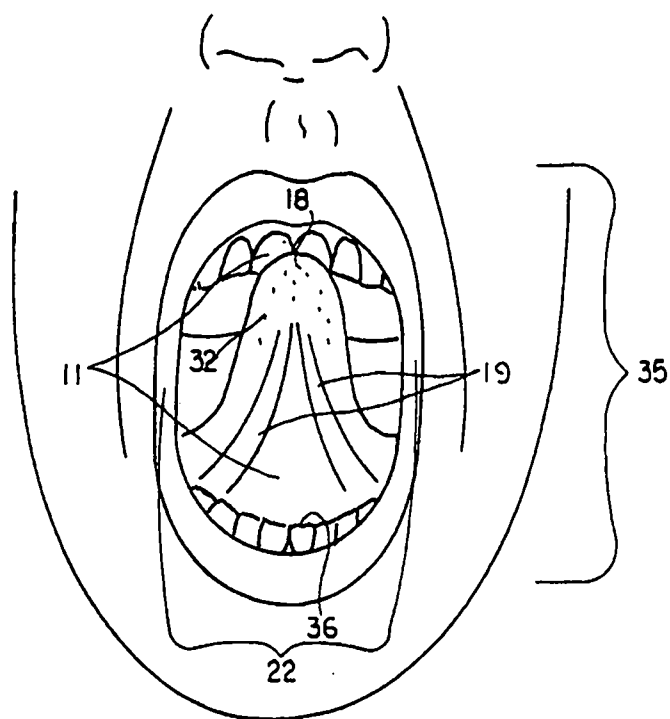
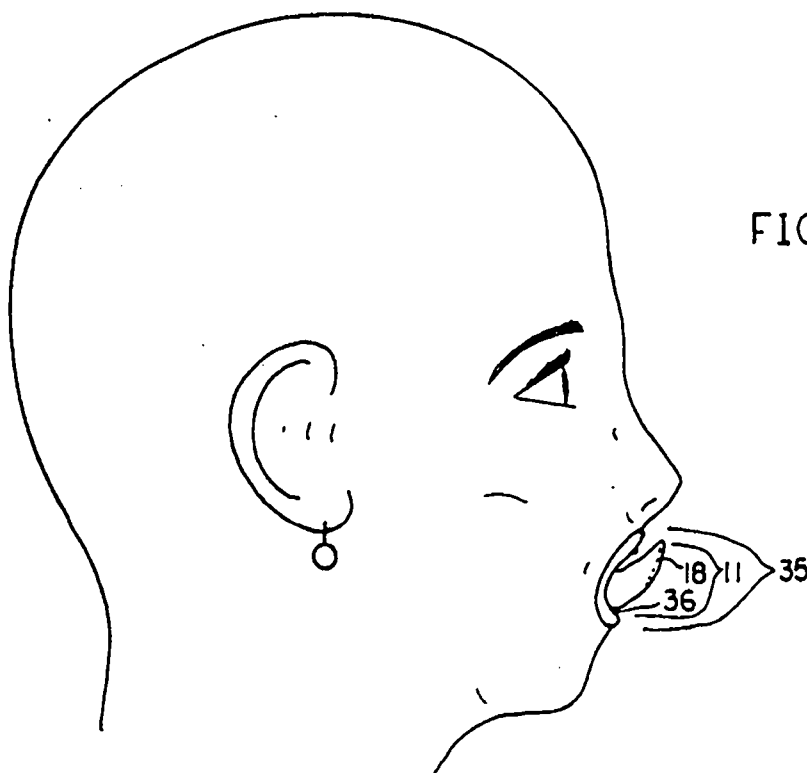
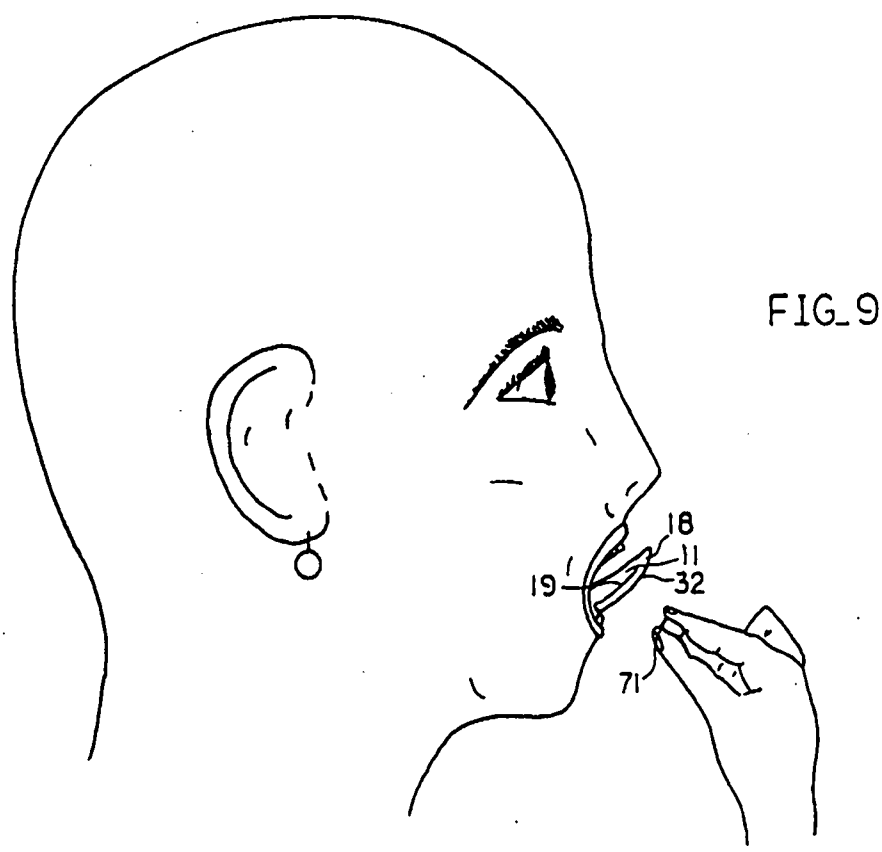
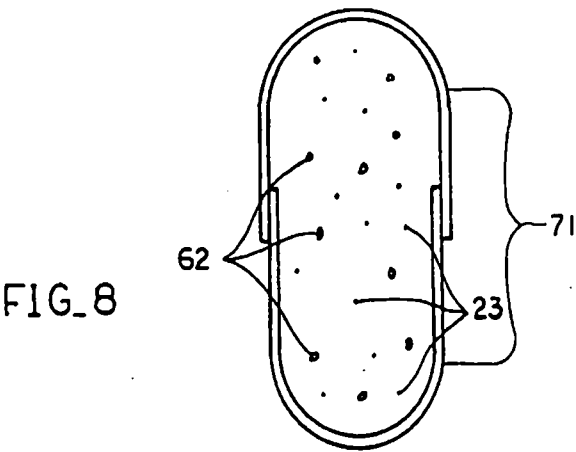


FIG. 7



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FORMULAE AND METHODS FOR SUBLINGUAL INGESTION OF NATURAL PROGESTERONE

SUMMARY OF THE INVENTION

The invention relates to pharmaceutical methods of preparing and dispensing the hormone Progesterone USP as well as a method of administering same.

Progesterone USP is a natural female body hormone. Under its influence the numerous minute glands which line the uterine cavity are transformed into secreting glands. This alteration is a part of the change which is essential to provide for the implantation of a fertilized ovum and for the continuing development of the placenta.

The present methods of effectively administering natural Progesterone (Progesterone USP) include suppositories, injection, and rectal suspension. Suppositories and injections are clearly the quickest method and, because of their direct access to the bloodstream, they are also the most effective.

The present invention relates to a form of Progesterone USP which is suitable for sublingual ingestion and methods for preparing Progesterone USP for sublingual ingestion. The invention also relates to the method of administering the medicine sublingually. Sublingual administration of the medicine, while resembling oral ingestion in comfort and convenience, has the added advantage of providing a direct path into the bloodstream. This results in a fast and efficient administration of the medicine.

Kincl (U.S. Pat. No. 3,193,457, Apr. 11, 1967) teaches the oral administration of various combinations of synthetic progestogens namely, dimethylprogesterone. It is therein pointed out that oral administration has advantages with respect to patients in a special state of sensitivity and mental stress. Kincl however, requires the dimethylprogesterone to pass through the stomach.

Kincl, however does not teach a method of the administration of Progesterone USP, as depicted in FIG. 1, but requires the use of other synthesized form of the hormone. Another such synthetic progestogen is depicted in FIG. 2.

Progesterone USP is the natural form of the hormone. As such it has the advantage of known compatibility with the human body which synthetic forms do not possess. This makes direct application to the bloodstream more feasible than with synthetic forms of the hormone. Additionally, there are certain "receptor sites" within the body which will not accept synthetic forms of Progesterone but will accept and process doses of Progesterone USP. These are the secretory cells of the mid-brain, the lining of the womb, the lungs, the eyes, the breasts, the liver, and the nasopharyngeal passages. Any application of Progesterone to these areas must be in the form of Progesterone USP.

A discussion of the processing of the sex hormones, including Progesterone USP within the body is helpful in summarizing the present invention and is schematically depicted in FIG. 3. Progesterone USP is manufactured by the ovaries and transmitted through the bloodstream to the secretory cells of the mid-brain. Here it is processed, along with the other various sex hormones into a variety of anterior pituitary hormones, such as prolactin, somatotrophic hormone, thyrotropin, adrenocorticotropin, follicle-stimulating hormone, and lutein-

izing hormone. It is these hormones which, in turn, stimulate healthy body metabolism.

The focus of the present invention is concerned with means and methods of delivering sex hormones, particularly Progesterone USP, to the secretory cells of the mid-brain, or other appropriate receptor sites for further body metabolism.

FIG. 4 depicts the path of medications, including Progesterone USP and other sex hormones, which are orally ingested. Entering the body through the mouth, they are next passed along to the stomach. From there they are processed, without significant chemical breakdown in the case of Progesterone USP, into the small intestines. From the small intestines they are transmitted, through the portal vein, into the liver. Within the liver, most medications, including Progesterone USP, are significantly broken down. For this reason oral ingestion of Progesterone USP is not effective in applications designed to treat deficiencies of the metabolic chain described in FIG. 3.

The sublingual mucosae, located on the underside of a person's tongue, provide expeditious entry into the general bloodstream of the person through the sublingual tributary veins. FIG. 5 schematically depicts the path taken by a medication entering the bloodstream at that point. Such a medication would first need to be absorbed through the sublingual mucosae into the sublingual veins. From there, the medication would be pumped into the right side of the heart and then into the lungs where the blood would be oxygenated. Progesterone USP would not be affected at this point. From there the oxygenated blood would carry the medication back into the left side of the heart and out through the systemic arteries for distribution throughout the body, including the secretory cells of the mid-brain and the other described receptor sites for Progesterone USP.

It would be advantageous to develop a form of Progesterone USP which could be administered without injections or suppositories in order to spare the related pain, inconvenience, or discomfort, yet would still permit quick and efficient entry into the bloodstream. Sublingual ingestion, or ingestion through the mucosae on the underside of a patient's tongue, would resolve this problem.

The present invention describes forms of Progesterone USP suitable for this purpose as well as the methods for milling and blending the Progesterone USP and methods of ingesting the Progesterone USP, so prepared. The invention further describes methods of sublingually administering the medication.

It is, then, an object of the invention to provide a form of pure natural Progesterone (Progesterone USP) which is capable of sublingual ingestion.

It is a further object of the invention to provide various methods of preparing a form of Progesterone USP which is capable of sublingual ingestion.

It is a further object of the invention to provide methods of sublingually administering Progesterone USP.

Further objects and advantages of the invention will become apparent during the detailed description and summary which follow.

BRIEF DESCRIPTION OF THE DRAWINGS

In having set out a background and summary of the invention as well as in the following detailed description of the invention, reference has been and will be made to the figures briefly described herein.

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FIG. 1 depicts the chemical structure of a typical synthetic progestogen.

FIG. 2 depicts the chemical structure of Progesterone USP.

FIG. 3 depicts the atomizer application of milled Progesterone USP according to the present invention.

FIG. 4 schematically depicts the path of medicines which are orally ingested.

FIG. 5 schematically depicts the path of medicines which are sublingually ingested.

FIG. 6 is a frontal view of the sublingual cavity.

FIG. 7 is a side view of the sublingual cavity.

FIG. 8 depicts a Progesterone USP capsule according to the present invention.

FIG. 9 depicts the application of a Progesterone USP capsule according to the present invention.

FIG. 10 depicts an atomizer with milled Progesterone USP according to the present invention.

DESCRIPTION OF THE PREFERRED EMBODIMENT

In providing a detailed description of the invention, direct reference will be made to the accompanying figures, numbered 3 and 6 through 10. It may prove helpful, however, to refer back to matters depicted in the previous figures for reference should difficulty be experienced in grasping the nature of the improvements and applications herein described.

Progesterone USP is marketed to the public through pharmacists as a prescription drug. Pharmacists typically receive Progesterone USP in a powder form. The present invention, in its preferred embodiments, is easily adapted from that form by further milling of the Progesterone USP with standard milling equipment. This process is well-known in the art and is not the subject of any claims herein. Consequently, no effort will be made to describe the milling process here, beyond providing data concerning the extent of milling required.

The sublingual area (11) of the mouth (35) is depicted in FIGS. 6 (front view) and 7 (lateral view). It generally comprises the area defined by the underside of the tongue (18) and the floor (36) of the mouth (35). The sublingual mucosae (32) are on the underside (37) of the tongue (18) along with the sublingual tributary veins (38).

Making reference now to FIGS. 8 and 9 the preparation of encapsulated Progesterone for sublingual administration and its sublingual administration will be described. First, Progesterone USP is milled as required such that no less than 75% of the total amount of it is in particle sizes which are less than five microns in diameter (23). The milled Progesterone USP (23) is then mixed with a salivic stimulator (62), such as citric acid. This mixture is then placed in a capsule (71), such as a "Lilly" brand capsule number 2, which is designed to readily dissolve when subjected to salivic juices in amounts normally found in the mouth.

In this form, the capsule can be placed under the patient's tongue (18) into the sublingual area (11). The capsule will quickly dissolve and the citric acid will stimulate the production of salivic fluid within the sublingual mucosae (32), maximizing the rate of sublingual absorption. The Progesterone USP will then be absorbed into the bloodstream (19) through the sublingual mucosae (32).

The concentration of salivic stimulator (62) should be within the range of 0.1 to 5 percent of the total amount of the mixture. There must be enough salivic stimulator

to ensure a presence of saliva on the sublingual mucosae (32). Too much saliva could result in significant amounts of the Progesterone USP (23) being carried away for oral ingestion of the Progesterone USP (23). This is precisely the event we seek to avoid because of the resulting premature breakdown of the medicine.

While citric acid is perhaps one of the more efficient salivic stimulators for use in this manner, a number of others, in the same relative concentrations, could be substituted. Some of the others which would be approved for use in this manner would be citric acid syrup, cherry juice, cherry juice syrup, cinnamon, cinnamon oil, clove oil, dextrose, ginger, glucose, lemon oil, lemon tincture, methyl salicylate, various orange extracts, various peppermint extracts, raspberry juice, raspberry syrup, various forms of saccharin, sorbitol, spearmint, spearmint oil, sucrose, syrup, various vanilla extracts, wild cherry syrup, and aspartame.

Alternate embodiments of the invention are depicted in FIGS. 3 and 10. In these embodiments of the invention, the Progesterone USP is prepared in such a manner as to permit application through an atomizer and the Progesterone USP is sublingually administered through an atomizer. Making reference first to FIG. 10, Progesterone USP is milled as required to facilitate its emulsification within fluids fit for human consumption. The fluids are used exclusively as an atomizer propellant vehicle. They must also be such that no chemical modification of the Progesterone USP will take place.

The milled Progesterone USP (91) emulsified within a suitable fluid (92) is then placed in an atomizer, generally (81), which is further adapted with an actuator (84). The actuator (84) is calibrated so as to accept only a precisely measured volume of the emulsified Progesterone USP (85) available for each application.

The operation of atomizers in providing a spray of fluids through a nozzle is well known in the art and not the subject of any claims herein. Consequently the functional principles of atomizers will be neither described nor depicted here, except to point out that the milling of the Progesterone USP must be sufficient to ensure that the particles of milled Progesterone emulsified within the fluid must be small enough to pass through the nozzle (86) of the atomizer (81). The nozzle (86) must be adapted to produce a spray the limits of which can easily be confined within the limits of a person's tongue (18).

Any concentration of Progesterone USP which permits its full emulsification within the fluid is acceptable. Natural oils, such as sesame seed, peanut, or vegetable oil are suitable for use as the propellant fluid.

One acceptable atomizer is manufactured by "Valois" of West Germany. It is equipped with a model number DF10 valve, which is 20 mm in length, an actuator model number 251 and an insert model number GP3. It should be noted that this method could be practiced with either a manually pumped or pressurized atomizer apparatus, so long as the pressure is sufficient to drive the emulsified Progesterone USP (85) through the atomizer and the atomizer was further equipped with an accurate actuator.

Making reference to FIG. 3, the administration of emulsified Progesterone USP (85) according to the present invention is described. The patient's tongue (18) is lifted and the blend (85) is sprayed against the underside (37) of the tongue, or into the sublingual area (11), again permitting immediate absorption into the bloodstream (19) through the sublingual mucosae (32). FIG.

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11 depicts the use of an atomizer (81) which is capable of precisely directing a band of spray (82) with radius (83) which is less than the width (22) of the patient's tongue (18).

Certain other modifications and embodiments of the invention as have been described herein may be made without departing from the spirit or the scope thereof and it is to be understood that the invention is to be limited only as defined in the appended claims.

What is claimed is:

1. A pharmaceutical formula for the administration of Progesterone USP said formula comprising;

Progesterone USP milled such that at least seventy-five percent of the total amount is composed of particle sizes of less than five microns diameter; said Progesterone USP being blended with a salivic stimulator in an amount adapted to stimulate sufficient saliva to dissolve said milled Progesterone USP but not enough to result in significant oral ingestion of Progesterone USP;

said mixture of Progesterone USP and salivic stimulator encapsulated within a soluble capsule.

2. The pharmaceutical formula described in claim 1 wherein said salivic stimulator is any combination of one or more of the flavoring materials selected from the list comprising;

citric acid, citric acid syrup, cherry juice, cherry juice syrup, cinnamon, cinnamon oil, clove oil, dextrose, ginger, glucose, lemon oil, lemon tincture, methyl salicylate, orange extract, peppermint extract, raspberry juice, raspberry syrup, saccharin, sorbitol, spearmint, spearmint oil, sucrose, syrup, vanilla extract, wild cherry syrup, and aspartame.

3. A method of preparing Progesterone USP such that it can be sublingually administered, said method comprising the steps of;

milling Progesterone USP such that at least seventy-five percent of said Progesterone USP is in particle size of no more than five microns diameter;

mixing said Progesterone USP with a salivic stimulation material; and

encapsulating said mixture within a capsule which is adapted to rapidly dissolve in the presence of saliva.

4. The pharmaceutical formula described in claim 1 wherein the salivic stimulator is citric acid in a concentration of between one-tenth percent and five percent of the entire formula.

5. The pharmaceutical formula described in claim 2 wherein the concentration of said salivic stimulators is

between one-tenth percent and five percent of the entire formula.

6. A method for sublingually administering Progesterone USP comprising the steps of,

suspending Progesterone USP within a fluid which is fit for human consumption and suitable for use as an atomizer propellant;

spraying said suspended Progesterone USP directly upon the underside of the tongue of the patient and permitting said suspended Progesterone USP to be sublingually absorbed into said patient's bloodstream.

7. A sublingually ingestible form of Progesterone USP, said form comprising,

Progesterone USP suspended within a suitable propellant fluid;

said suitable propellant fluid being fit for human consumption and of such density that said suspended Progesterone USP can be passed through an atomizer nozzle.

8. A method of administering Progesterone USP, comprising the steps of;

milling Progesterone USP such that at least seventy-five percent of said Progesterone USP is in particle size of no more than five microns diameter;

mixing said Progesterone USP with a salivic stimulation material;

encapsulating said mixture within a capsule which is adapted to rapidly dissolve in the presence of saliva; and

placing said encapsulated Progesterone USP between the lower surface of the patient's mouth and the underside of the patient's tongue.

9. A method of sublingually administering Progesterone USP comprising the steps of;

milling the Progesterone USP to a size small enough to pass through the nozzle of an atomizer;

suspending said milled Progesterone USP within a suitable fluid which is suitable for human consumption and will neither break down nor change the chemical nature of said suspended Progesterone USP; and

placing said suspended Progesterone USP into an atomizer; and

spraying said suspended Progesterone USP upon the underside of the patient's tongue.

10. The method described in claim 9, in which said suitable fluid is peanut oil.

11. The method described in claim 9, in which said suitable fluid is sesame seed oil.

12. The method described in claim 9, in which said suitable fluid is vegetable oil.

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