

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO



No. 06-120701 - 00003-0000

Fecha: 2008-06-13 17:39:05 Dep: 2020 NUECREACION
Tra: 11 PATENTHYII Ene: 379 FASE-NACIONAL
Act: 538 PETICION Folios: 3

jllcco@lloredacamacho.com

Señor

SUPERINTENDENTE DE INDUSTRIA Y COMERCIO

DIVISION DE NUEVAS CREACIONES

E. _____ S. _____ D. _____

Re: **P1.-GLAXOSMITHKLINE BIOLOGICALS S.A.-Solicitud de Patente de Invención en Fase Nacional PCT para "VACUNA CONTRA HPV16 Y HPV18 Y AL MENOS OTRO TIPO DE VPH SELECCIONADO DE VPH 31, 45 O 52".-Clase A61K 39/12.-Expediente número 06-120.701.-**

1. Me refiero a mi memorial de 2 de Abril de 2007.
2. De conformidad con lo dispuesto en el Artículo 5.2.12 de la Circular Externa No. 03 de 28 de Abril de 2003, y estando dentro del plazo de seis meses contados desde la fecha de la publicación de la solicitud en la Gaceta de la Propiedad Industrial número **583** de 18 de Diciembre de 2007, solicito a ese Despacho proceder a realizar el examen de patentabilidad de la presente invención.
3. Por otro lado, atentamente me permito manifestar a usted que en el extracto de la solicitud arriba nombrada, publicado en la Gaceta de la Propiedad Industrial número **583** de 18 de Diciembre de 2007, página 77, la fecha de la publicación internacional aparece publicada en forma equivocada, la fecha de la publicación internacional correcta es **29 de Diciembre** de 2005, como se indicó en el petitorio y el extracto de la solicitud.
4. En vista de lo anterior, muy comedidamente solicito a usted que las aclaraciones correspondientes sean incluidas en la Fe de Erratas de la próxima Gaceta de la Propiedad Industrial.

5. Me permito recordar a usted, que tal como se informó en el Petitorio de la invención, la presente solicitud corresponde a Capítulo II.
6. Anexo me permito presentar recibo número 08-45.194, expedido por la Superintendencia de Industria y Comercio, que acredita el pago de la tasa que dicho examen causa.
7. Ruego a usted tomar atenta nota de lo anterior y proceder de conformidad.

Señor Superintendente,

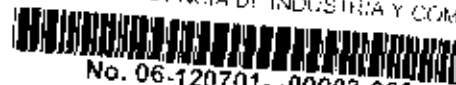


ALICIA LLOREDA RICAURTE

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SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO



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Fecha: 2008-06-13 17:39:05 Dep: 2020 NÚMERO DE CREACIÓN:
Tit. 11 PATENTE DE MARCA EVO 379 FASE NACIONAL II
Act. 538 PETICIÓN Folios: 3

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO

NIT : 800174089-2

RECIBO OFICIAL DE CAJA : 08 - 45,194
: JUNIO 6 DE 2008

==== CONSIGNACION =====

DEPOSITANTE	TIPO PAGO	BANCO	CUENTA	Nº. PAGO	FECHA PAGO	Vr. PAGO
LEUREZA Y CIA S. A	CONSIGNACION	BANCO DE BOGOTA	62754327	2756025	06/06/2008	8,619,300.00

==== C O N C E P T O =====

RENTISTICO	CONCEPTO	TOTAL CONCEPTO
50005-01-01 SOLICITUDES	114 PTO CAP-II EXAMEN DE PATENTABILIDAD	210,000.00
	TOTAL :	210,000.00

SON: OCHOCIENTOS DIEZ MIL PESOS

RESPONSABLE : _____

RECIBO DE CAJA APLICADO AL EXPEDIENTE No. _____

JOSE LLOREDA CAMACHO & CO.

ABOGADOS

Calle 72 No. 5-83 Piso 6, Bogotá, D.C. Tel: 8069700 Fax 8069701

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SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO



No. 06-120701.-00004-0000

Señor

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DIVISION DE NUEVAS CREACIONES

Fecha: 2011-07-19 17:19:44 Dep. 2020 DIR.NUEVASOR
Tra. 11. PATENTEYII Eva: 379 FASENACIONALI
Act. 339 COMUNICACIÓN Folios: 1

E. S. D.

Re: PI.-GLAXOSMITHKLINE BIOLOGICALS S.A.-Solicitud de Patente de Invención para "VACUNA CONTRA HPV16 Y HPV18 Y AL MENOS OTRO TIPO DE VPH SELECCIONADO DE VPH 31, 45 O 52."-Clase A61K 39/12.- Expediente número 06-120.701.-

1. Me refiero a mi memorial de 13 de Junio de 2008.
2. Atentamente ruego a usted, se sirva pronunciarse en relación con el examen de fondo de la solicitud arriba nombrada, ya que desde el 18 de Diciembre de 2007, esto hace casi cuatro (4) años que fue publicada la solicitud en la Gaceta 583, y en su debido momento mediante memorial de 13 de Junio de 2008, se solicitó y pago la tasa correspondiente al examen de patentabilidad sin que hasta el momento haya sido adelantado dicho trámite.
3. Cabe anotar que esta demora en el trámite acarrea grave perjuicio para mi representado.

Señor Superintendente,

ALICIA LLOREDA RICAURTE

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DIRECCIÓN DE NUEVAS CREACIONES

RADICACIÓN EXPEDIENTE No. 06-120701

EL EXTRACTO DE LA PRESENTE SOLICITUD DE **PATENTE DE INVENCION**
FUE PUBLICADO EN LA GACETA DE PROPIEDAD INDUSTRIAL No **583** DE
FECHA **18 DE DICIEMBRE DE 2007** EL TÉRMINO HÁBIL SEÑALADO POR LA
LEY EXPIRA EL **14 DE MARZO DE 2008**

NOTA: Dentro del plazo de los seis (6) meses siguientes a partir de la fecha de publicación en la gaceta de Propiedad Industrial, deberá solicitar y cancelar el examen de patentabilidad so pena de que la solicitud caiga en abandono, de acuerdo con lo establecido en el Artículo 44 de la Decisión 486/2000.

SE PRESENTARON OPOSICIONES POR PARTE DE TERCEROS.

SI _____ NO X

(43) World Intellectual Property Organization
International Bureau



(44) International Publication Date
25 September 2003 (25.09.2003)

PCT

(11) International Publication Number
WO 03/077942 A2

(51) International Patent Classification:

(86) Designated States (optional):

(71) International Application Number: 497,414 (90.0000)

AZ, BA, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EA, GB, GR, HU, IL, IN, JP, KR, KZ, LC, MG, MK, MN, MW, MX, NO, NZ, OM, PE, PL, PT, RO, RU, SC, SD, SE, SG, SK, SI, TJ, TM, TR, TT, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW

(72) International Filing Date: 17 March 2003 (17.03.2003)

AM, AR, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EA, GB, GR, HU, IL, IN, JP, KR, KZ, LC, MG, MK, MN, MW, MX, NO, NZ, OM, PE, PL, PT, RO, RU, SC, SD, SE, SG, SK, SI, TJ, TM, TR, TT, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW

(73) Filing Language: English

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(20) Priority Date: 18 March 2002 (18.03.2002)

(84) Designated States (optional):

(71) Applicant for all designated States except USA: CLAX-OSMITH LINE BROOKING & SONS (PVT) LTD., 100, South Lakes Boulevard, one de l'Union Rd, H-1130 Bavi, Beirut (LB)

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(72) Inventor and

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(73) Inventor(s) Applicant for (US only): WEITENSON, E. M., 100, South Lakes Boulevard, one de l'Union Rd, H-1130 Bavi, Beirut (LB)

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(74) Agent: GIBBINS, Peter, John, 100, South Lakes Boulevard, one de l'Union Rd, H-1130 Bavi, Beirut (LB)

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Viral antigens

The present invention relates to vaccines against HPV. In particular the invention relates to vaccines comprising virus like particles (VLPs), especially virus like particles comprising proteins from human papilloma virus (HPV).

Papillomaviruses are small DNA tumour viruses, which are highly species specific. So far, over 100 individual human papillomaviruses (HPV) genotypes have been described. HPVs are generally specific either for the skin (e.g. HPV-1 and -2) or mucosal surfaces (e.g. HPV-6 and -11) and usually cause benign tumours (warts) that persist for several months or years. Such benign tumours may be distressing for the individuals concerned but tend not to be life threatening, with a few exceptions.

Some HPVs are also associated with cancers. The strongest positive association between an HPV and human cancer is that which exists between HPV-16 and HPV-18 and cervical carcinoma. Cervical cancer is the most common malignancy in developing countries, with about 500,000 new cases occurring in the world each year. It is now technically feasible to actively combat primary HPV-16 infections, and even established HPV-16-containing cancers, using vaccines. For a review on the prospects for prophylactic and therapeutic vaccination against HPV-16 see Cason J., Clin. Immunother. 1994, 1(4) 293-306 and Hagensee M. H., Infections in Medicine 1997 14(7) 555-556, 559-564.

Although minor variations do occur, all HPVs genomes described have at least eight early genes, E1 to E8 and two late genes L1 and L2. In addition, an upstream regulatory region harbours the regulatory sequences which appear to control most transcriptional events of the HPV genome.

HPV L1 based vaccines are disclosed in WO94/00152, WO94/20137, WO93/02184 and WO94/05792. Such a vaccine can comprise the L1 antigen as a monomer, a cosubstrate or a virus like particle. Methods for the preparation of VLPs are well known in the art, and include VLP disassembly-reassembly approaches to provide enhanced immunogenicity, for example as described in WO99/13056 and US6245568. Such particles may additionally comprise L2 proteins. L2 based vaccines are

described, for example, in WO93/00436. Other HPV vaccines are based on the L1 proteins, such as L1 or fusion proteins such as L2-E7.

Despite the work on IIPV vaccines there is still no broadly effective vaccine against cervical cancer.

The present invention relates to an improved vaccine against human papilloma virus.

In a first aspect the present invention relates to a vaccine composition comprising VLPs containing L1 proteins or functional L1 protein derivatives from HPV 16, HPV 18, HPV 31 and HPV 45.

The invention also relates to a method of vaccine production, the method comprising combining VLPs containing L1 proteins or functional L1 protein derivatives from HPV 16, HPV 18, HPV 31 and HPV 45.

The invention further relates to use of a mixture of VLPs containing L1 proteins or functional L1 protein derivatives from HPV 16, HPV 18, HPV 31 and HPV 45 in the preparation of a vaccine for the prevention of cervical cancer.

The invention further relates to a method of preventing cervical cancer, the method comprising delivering to an individual at risk of cervical cancer an effective amount of a vaccine as described above, such as a vaccine comprising a mixture of HPV 16, HPV 18, HPV31 and HPV 45 VLPs.

The VLPs of the present invention can be formed from either the full length HPV L1 protein or certain L1 derivatives using standard methods in the art, for example as disclosed in WO99/13056 incorporated herein by reference.

It is preferred that the L1 protein used to form the VLP is a truncated L1 protein. Preferably at least one of the VLPs comprises a truncated L1 protein, and preferably all the L1 proteins in the combination vaccine are truncated L1 proteins. Preferably the truncation removes a nuclear localisation signal. Preferably the truncation is a C-terminal truncation. Preferably the C-terminal truncation removes fewer than 50

amino acids, more preferably fewer than 40 amino acids. Most preferably the C-terminal truncation removes 34 amino acids from HPV 16 and 35 amino acids from HPV 18.

Truncated L1 proteins are suitably functional L1 protein derivatives. Functional L1 protein derivatives are capable of raising an immune response (if necessary, when suitably adjuvanted), said immune response being capable of recognising a VLP consisting of the full length L1 protein and/or the HPV type from which the L1 protein was derived.

VLPs of the invention may also comprise other types of functional protein derivatives, including mutants of the full length or truncated HPV L1 proteins such as deletion, substitution, or insertion mutants. Suitable derivatives also include codon optimised sequences. The L1 protein or derivative may also be a fusion protein, such as the fusion of the L1 protein with L2 or an early protein. Preferably fusion proteins comprise proteins from only one HPV genotype. VLPs made from chimeric L1 proteins in which L1 proteins from one genotype are linked to L1 proteins from other genotypes are not preferred.

The L1 protein or functional protein derivative is suitably able to form a VLP, and VLP formation can be assessed by standard techniques such as, for example, electron microscopy and dynamic laser light scattering.

Preferably the polydispersity of the VLPs is less than 0.15, most preferably less than 0.1 and more preferably less than 0.08 when measured using a Malvern Zetasizer 3000HS under conditions as described herein.

Use of the term 'protein' or reference to a specific protein e.g. 'L1' is hereinafter taken to include reference to functional protein derivatives, unless otherwise indicated or obviously apparent from the context.

In a preferred aspect of the invention the vaccine of the invention has only four VLP types - HPV 16, HPV 18, HPV 31 and HPV 45 VLPs. Preferably the VLPs are L1 only VLPs from each of these 4 genotypes.

Alternatively, and most preferred, the vaccine comprises an additional HPV valency, making a pentavalent vaccine. Preferably the additional valency is a VLP comprising an L1 protein or functional derivative, as above, from one of HPV 52, 53, 58, 33, 35, 56, and 59. Preferably the 5th genotype is HPV 33 when the vaccine is for use in South America or HPV 52, 53 or 58 when the vaccine is for use in Asia.

The present invention also extends to vaccines comprising 2 or more additional valencies, to provide a vaccine with 6 or more genotypes.

In one preferred embodiment the combination excludes VLPs from HPV 6a, 6b or HPV 11 genotypes.

Preferably the vaccine of the invention is at least 55% effective in preventing cervical cancer, more preferably 60%, 65%, 70%, 75% preferably 80% or even more effective in prevention of cervical cancer. For the avoidance of doubt, % efficacy in prevention of cervical cancer means protection against all cervical cancer induced by HPV infection, and not just protection against cancer caused by one genotype. Prevention may be suitably assessed over 1 year post initial vaccination, although preferred vaccines are equally effective over 2, 3, 4, 5 or more years. The % efficacy can be increased by selecting appropriate HPV genotypes to target the vaccine formulation to specific geographical areas.

Preferably the combination of VLPs within the vaccine does not reduce the immunogenicity of each VLP type. In particular it is preferred that there is no interference between HPV VLPs in the combination of the invention, such that the combined VLP vaccine of the invention is able to offer effective protection against infection by each HPV genotype represented in the vaccine. Suitably the immune response against a given VLP type in the combination is at least 50 % of the immune response of that same VLP type when measured individually, preferably 100% or substantially 100%. For responses to the HPV 16 and HPV 18 VLPs, the combined vaccine of the invention preferably stimulates an immune response which is at least 50% of that provided by a combined HPV 16 / HPV 18 VLP vaccine. Suitably the immune response generated by the vaccine of the invention is at a level in which the

protective effect of each VLP type is still seen. The immune response may suitably be measured, for example, by antibody responses, as illustrated herein.

The vaccine of the invention may be used to treat or prevent HPV infection and/or disease. For example the vaccine may be used therapeutically to reduce viral load and/or infections that lead to cervical carcinoma or CIN III sequelae. The invention thus relates to use of the vaccine of the invention in the therapeutic treatment of diseases related to HPV infection and in prophylaxis of infection or disease. The invention also relates to use of the VLP combination of the invention in generation of an immune response against HPV 16, 18, 31 and 45.

The vaccine of the invention may optionally be formulated with VLPs which provide protection against genital warts, such as VLPs containing L1 protein from HPV 6a, 6b and/or HPV 11 genotypes.

Preferably the VLPs comprise the HPV L1 protein only and no L2 protein or protein fragment.

Vaccines of the invention may comprise other proteins or protein fragments in addition to the L1 protein or derivative. Proteins/peptides may be delivered in conjugate form with the L1 protein in the VLP, encapsulated within a VLP or co-formulated in a mixture with the VLP's. Other proteins or peptides may also be co-administered with the vaccine of the invention.

In one aspect the vaccine comprises an HPV L2 protein or L2 derivative such as an L2 peptide, for example as disclosed in K. Kawana et al Vaccine 19, (2001) p1496-1502, incorporated herein by reference. In a further preferred embodiment the vaccine of the invention may be formulated with HPV early antigens such as E1, E2, E3, E4, E5, E6, E7, E8 or immunologically active derivatives thereof. When delivered in conjugate form it is preferable to utilise an immunogenic fragment of about 30-60 amino acids of the early antigen.

Optionally the vaccine may also be formulated or co-administered with non-HPV antigens. Suitably these antigens can provide protection against other diseases, most

preferably sexually transmitted diseases such as herpes simplex virus, chlamydia and HIV. We particularly prefer that the vaccine comprises gD or a truncate thereof from HSV, preferably the gD21 protein as described in WO 99/45957. In this way the vaccine provides protection against both HPV and HSV. Preferred HIV antigens are described in WO/9916884 and WO/0154719.

The present invention generally relates to a mixture of VLPs containing capsid proteins from HPV16, 18, 31 and 45, such as L1-only VLPs.

In a particularly preferred embodiment, the invention provides a vaccine comprising a mixture of HPV 16 VLPs, HPV 18 VLPs, HPV 31 VLPs and HPV 45 VLPs.

Reference herein to "HPV 16 VLP", for example, is a reference to an L1 VLP wherein the L1 protein or L1 derivative is from HPV 16. The same nomenclature principle applies, by extension, to other VLPs described herein, such as HPV 18, HPV 31 and HPV 45 VLPs.

Preferably each VLP contains L1 protein from only 1 HPV genotype. Such a vaccine may be formulated by production of individual VLPs from HPV 16, 18, 31 and 45, followed by combination of such VLPs. Preferably there are no other HPV proteins in the VLP other than L1.

Also preferred are VLPs containing proteins from only one HPV genotype, such as VLPs with L1 and L2 from HPV 16.

However, in an alternative embodiment of the invention, the VLPs may be mixed VLPs, a mixed VLP comprising L1 protein from one genotype in combination with L1 protein from a second genotype, wherein the different L1 proteins are not chimeric L1 proteins, but associate together within the same capsid structure to form immunogenic VLPs.

Preferred combinations include any permutation of genotypes 16, 18, 31 and 45 - for example, the invention may comprise a mixed HPV 16/HPV 18 VLP in combination with a mixed HPV 31/HPV 45 VLP, or mixed 16/31 VLPs in combination with mixed

18/45 VLPs. Combinations of more than 2 L1 genotypes within 1 VLP are also contemplated.

Mixed VLPs may be produced by separate expression of the individual L1 proteins followed by combination to form VLPs, as exemplified herein. Alternatively multiple L1 proteins may be expressed within the same cell, from one or more DNA constructs. For example, multiple DNA constructs may be transformed or transfected into host cells, each vector encoding a different L1 protein. Alternatively a single vector having multiple L1 genes, controlled by a shared promoter or multiple individual promoters, may be used. IRES elements may also be incorporated into the vector, where appropriate. Using such expression strategies the co-expressed L1 proteins may be co-purified for subsequent VLP formation, or may spontaneously form mixed VLPs which can then be purified.

Where mixed VLPs are used, a preferred process for mixed VLP production comprises preparation of VLP L1 proteins or derivatives, such as L1 proteins, from different papillomavirus genotypes, mixing the proteins if necessary and assembly of the proteins to produce mixed VLPs. The L1 proteins may be in the form of a crude extract, be partially purified or purified prior to mixing. Preferably the proteins are at least partially purified before being combined. Optionally, further purification of the mixed VLPs may be carried out after assembly. Where additional antigens are used, then these may be added where appropriate.

In one embodiment mixed VLPs may be made by disassembly of 2 or more VLPs, followed by combination of the disassembled VLP components at any suitable point prior to reassembly. This approach is suitable when VLPs spontaneously form when the L1 protein is expressed, as occurs for example, in some yeast strains. Where the expression of the L1 protein does not lead to spontaneous VLP formation, preparations of L1 proteins or capsomers may be combined before assembly into VLPs.

Assembly of VLPs is generally achieved by removal of a reducing agent. As such, in mixed VLP production, the mixing of proteins preferably takes place prior to the removal of a reducing agent from the mixture of proteins. Preferably the production

of mixed VLPs comprises the step of mixed VLP formation from a mixture of dissociated L1 proteins by removal of a reducing agent from the mixture under conditions that allow VLPs to form.

Preferably the reassembly process results from removal of a reducing agent such as β -mercaptoethanol.

It is, however, known that VLP formation is dependent upon pH, metal ions and salinity as well as the presence of a reducing agent. As such, under certain circumstances, it may be envisaged that VLPs might form in the presence of a reducing agent. It is only important to the invention that mixing of the proteins from different genotypes occurs prior to the change in environmental condition that allows the mixed VLPs to form, whether this is pH, metal ions, salinity, reducing environment or combination of these.

Where mixed VLPs are used, preferably the components of the VLPs are mixed in the proportions in which they are desired in the final mixed VLP. For example, a mixture of the same amount of a partially purified L1 protein from HPV 16 and HPV 18 provides a mixed VLP with approximately equal amounts of each protein.

Vaccine solutions comprising mixed VLPs may be stabilised by compositions known in the art, such as those of WO 98/44944, WO0045541, incorporated herein by reference.

For all vaccines of the invention, it is preferred that the vaccine is used for the vaccination of adolescent girls aged 10-15, preferably 10-13 years. The vaccine may also be administered to women following an abnormal pap smear or after surgery following removal of a lesion caused by HPV.

Preferably the vaccine is delivered in a 2 or 3 dose regime, for example in a 0, 1 month regime or 0, 1 and 6 month regime respectively. Suitably the vaccination regime incorporates a booster injection after 5 to 10 years, preferably 10 years.

Preferably the vaccine is a liquid vaccine formulation, although the vaccine may be lyophilised and reconstituted prior to administration.

The vaccines of the invention may also comprise adjuvants in combination with the VLPs. Suitably the VLPs of the invention are used in combination with aluminium, and are suitably adsorbed or partially adsorbed onto aluminium adjuvants. Also preferred are adjuvants which stimulate a Th1 type response such as 3D-MPL or QS21. Suitably the adjuvant is an aluminium salt, preferably in combination with 3D-MPL, such as aluminium phosphate and 3D-MPL.

A preferred adjuvant is aluminium hydroxide, with the combination of aluminium hydroxide with 3D-MPL especially preferred.

When VLPs are adsorbed on to aluminium containing adjuvants, the adjuvant is preferably added before mixing of the VLPs to form the final vaccine product.

The vaccine may also comprise aluminium or an aluminium compound as a stabiliser, and the present invention also relates to a stabilised combination vaccine wherein the VLPs are adsorbed onto an aluminium salt. Suitably the VLPs are more stable over time after adsorption onto an aluminium salt than in the absence of aluminium. Preferably stabilised VLPs are obtained or obtainable by methods according to example 1 section C3.

The vaccines of the invention may be provided by any of a variety of routes such as oral, topical, subcutaneous, mucosal (typically intravaginal), intravenous, intramuscular, intranasal, sublingual, intradermal and via suppository. Intramuscular and intradermal delivery are preferred.

The dosage of VLP and other proteins will vary with the condition, sex, age and weight of the individual, the administration route and HPV of the vaccine. The quantity may also be varied with the number of VLP types. Suitably the delivery is of an amount of VLP suitable to generate an immunologically protective response. Suitably each vaccine dose comprises 1-100 μ g of each VLP, preferably 5-80 μ g.

more preferably 5- 30 µg each VLP, most preferably 5- 20 µg of each VLP with 5µg, 6µg, 10µg, 15 µg or 20µg especially preferred.

The multivalent vaccine of the present invention is suitably produced by combining purified L1 VLPs. Methods for the production of L1 VLPs are well known in the art, and include for example methods given in WO9531532, WO9615247, WO00/69671 and US5885326, the whole contents of which are incorporated herein.

Suitably the VLPs of the invention are made by disassembly and reassembly of VLPs, to provide homogenous and pure VLPs. Examples of suitable processes are given in WO057906, US6245568 and WO9913056.

Preferably the VLPs are prepared from insect cells such as Sf9 or Hi 5 cells, although any suitable cells such as E. coli or yeast cells, for example, *S. cerevisiae*, *S. pombe* or *Pichia* sp. may also be used.

Preferably the purification of VLPs after L1 expression includes one or more of the steps of anion exchange chromatography (Di methyl amino ethyl – DMAE), anion exchange chromatography (tri methyl amino ethyl – TMAE), hydroxyapatite chromatography, filtration such as nanometric filtration or ultrafiltration, or hydrophobic interaction chromatography. Preferably at least one anion exchange step is performed during purification, and more preferably 2 anion exchange steps are used. Preferably at least one anion exchange purification step is performed prior to mixing the proteins. Optionally a UV irradiation step may be employed.

For the avoidance of doubt, the entire teaching of all documents referred to herein is incorporated by reference.

The present invention is illustrated by the following non-limiting Examples and Figures, wherein:

Figure 1 illustrates mixed VLPs in comparison with HPV 16 VLPs as assessed by EM;

Figures 2 and 3 illustrate size distribution of mixed VLPs;

Figure 4 illustrates antibody responses against VLP 16 in a mixed HPV 16, 18, 31, 45 combination vaccine vs. an HPV 16 control;

Figure 5 illustrates antibody responses against VLP 18 in a mixed HPV 16, 18, 31, 45 combination vaccine vs. an HPV 18 control;

Figure 6 illustrates antibody responses against VLP 31 in a mixed HPV 16, 18, 31, 45 combination vaccine vs. an HPV 31 control; and

Figure 7 illustrates antibody responses against VLP 45 in a mixed HPV 16, 18, 31, 45 combination vaccine vs. an HPV 45 control.

Example 1

The combination of HPV 16 and HPV 18 L1 VLPs is detailed herein. L1 proteins from other HPV genotypes may be readily produced by similar methods, already known in the art.

A Preparation of HPV 16/18 L1 VLPs

Production of HPV 16 and HPV 18 VLPs was carried out using standard protocols (for example, see WO9913056). HPV 16/18 proteins were expressed in *Trichoplusia ni* (High Five™) cells (at a density of ~ 350000 cells/ml) infected with recombinant Baculovirus (MOI of 0.3) encoding the HPV 16 or 18 L1 gene of interest. Cells were harvested approximately 72 hours post infection.

B Cell harvest / antigen extraction

The antigen (L1-16/18) was extracted from H15 cells in a three step process of concentration, extraction, clarification. The concentration step consists removes up to 90% of the culture medium, and was performed by tangential flow filtration. The extraction step was performed with a hypotonic buffer (Tris 20mM, pH 8.5). A volume equal to the culture volume was used to perform the extraction. A contact time of minimum half an hour under smooth agitation was used. The clarification was performed by tangential flow filtration.

C Purification

The purification process was carried out at room temperature. β -mercaptoethanol (4% w/w) was added to the extract in order to disassemble the VLP's into capsomers, for both antigens, 1.1-16/18. Glycerol was added up to a concentration of w/w 10% just prior to the addition of β -mercaptoethanol.

All buffers used were filtered on 0.22 μ m filters prior to storage at 2°C-8°C. Prior to each purification run, gel matrixes are sanitised and equilibrated with appropriate buffer before sample loading.

Purification regimes are given for the separate purification of L1 from both HPV 16 and 18. These schemes are broadly similar, and involve the steps of:

Anion exchange chromatography (Di methyl amino ethyl - DMAE),

Anion exchange chromatography (tri methyl amino ethyl - TMAE),

Hydroxyapatite chromatography,

Nanometric Filtration (Planova),

Ultrafiltration,

Hydrophobic interaction chromatography (using Octyl Sepharose) for HPV 18 or

Anion exchange chromatography (DEAE) for HPV 16; and

Sterile filtration.

Specifically:

C1 Purification of L1-18 antigen

Anion exchange chromatography DMAE

The clarified extract (protein at a concentration of ~ 1 μ g/ml, with the L1 protein at ~ 150 μ g/ml) is applied to an anion exchange column (Di Methyl Amino Ethyl).

Elution is performed with (Tris 20mM | NaCl 200mM | 4% β -mercaptoethanol BME) buffer, pH 7.9 $\pm$ 0.2. The antigen is eluted in approximately 5 column volumes and the elution profile is monitored at 280 nm.

Anion exchange chromatography TMAE

The eluate of the first step is diluted with 1 volume of H₂O/BME 4%. The diluted eluate is then applied to a second anion exchange column (Tri Methyl Amino Ethyl). Elution is performed with (20mM Tris | NaCl 200mM | 4%BME) buffer, pH 7.9 $\pm$ 0.2. The antigen is eluted in approximately 4 column volumes and the elution profile is monitored at 280 nm.

Hydroxyapatite chromatography

The eluate of the TMAE step is applied to a hydroxyapatite (HA) column.

After sample application, the gel is eluted with approximately 2.5 column volumes of (NaH₂PO₄ 100mM | NaCl 30mM | 4%BME) buffer, pH 6.0 $\pm$ 0.2.

Nanometric filtration (Planova)

The HA eluate is diluted in order to reach the following conditions: (NaH₂PO₄ 25mM | NaCl 10mM | 4%BME) buffer, pH 7.5 $\pm$ 0.2.

Then it is filtered successively on a 0.2 μ m prefilter and on a Planova 15N filter of 0.12 m². The filtration is performed at constant pressure 200 mbar $\pm$ 20 mbar.

Ultrafiltration

The ultrafiltration is performed with a tangential flow ultrafiltration system equipped with polyethersulfone membranes (Centramate cassette 0.1 m², 100kD).

The Planova eluate is treated to reach the following conditions: (NaH₂PO₄ 100mM | NaCl 30mM | 4%BME), pH 6.0 $\pm$ 0.2; then it is loaded in the system, concentrated 5 fold and dia-filtrated with continuous injection of ~10 starting volumes of (NaH₂PO₄ 20mM | NaCl 500mM) buffer, pH 6.0 $\pm$ 0.2.

Hydrophobic interaction chromatography (Octyl Sepharose)

The ultrafiltration permeate is applied to an Octyl Sepharose column.

This chromatography step is run in the negative mode with approximately 5 column volumes of (Na₂PO₄ 20mM | NaCl 500mM) buffer, pH 6.0 $\pm$ 0.2.

Sterile filtration

The purified L1-18 antigen solution is sterilised by filtration on a 0.22 μ m membrane.

C2 Purification of L1-16 antigen

Anion exchange chromatography DMAE

The clarified extract is applied to an anion exchange column (Di Methyl Amino Ethyl).

Elution is performed with (Tris 20mM | NaCl 180mM | 4%BME) buffer, pH 7.9 ± 0.2.

The antigen is eluted in approximately 4 column volumes and the elution profile is monitored at 280 nm.

Anion exchange chromatography TMAE

The eluate of the first step is diluted with 1 volume of H₂O/BME 4%. The diluted eluate is then applied to a second anion exchange column (Tri Methyl Amino Ethyl).

Elution is performed with (20mM Tris | NaCl 180mM | 4%BME) buffer, pH 7.9 ± 0.2.

The antigen is eluted in approximately 5 column volumes and the elution profile is monitored at 280 nm.

Hydroxyapatite chromatography (HA)

The eluate of the TMAE step is applied to a HA column.

After sample application, the gel is eluted with approximately 3 column volumes of (NaH₂PO₄ 100mM | NaCl 30mM | 4%BME) buffer, pH 6.0 ± 0.2.

Nanometric filtration (Planova)

The HA eluate is diluted in order to reach the following conditions: (NaH₂PO₄ 25mM | NaCl 10mM | 4%BME) buffer, pH 7.5 ± 0.2.

Then it is filtered successively on a 0.2 µm prefilter and on a Planova 15N filter of 0.12 m². The filtration is performed at constant pressure 200 mbar ± 20 mbar.

Ultrafiltration

The ultrafiltration is performed with a tangential flow ultrafiltration system equipped with polyethersulfone membranes (Centramate cassette 0.1 m², 100kD).

The Planova eluate is treated to reach the following conditions: (NaH₂PO₄ 100mM | NaCl 30mM | 4%BME), pH 6.0 ± 0.2; then it is loaded in the system,

concentrated 5 fold and dia-filtered with continuous rejection of ~10 starting volumes of (NaH₂PO₄ 20mM | NaCl 500mM) buffer, pH 6.0 ± 0.2.

Anion exchange chromatography DEAE

The ultrafiltration eluate is adjusted to the conductivity of the equilibrium buffer, (Na₂PO₄ 20mM | NaCl 250mM), pH 6.0 ± 0.2 and applied on an anion exchange column (Di Ethyl Amino Ethyl).

Elution is performed with (NaH₂PO₄ 20mM | NaCl 500mM) buffer, pH 6.0 ± 0.2. The antigen is eluted in approximately 3 column volumes and the elution profile is monitored at 280 nm.

Sterile filtration

The purified L1-16 antigen solution is sterilised by filtration on a 0.22 µm membrane.

C3

Each VLP type is adsorbed independently to produce a concentrated adsorbed monovalent.

Preparation of VLP16 concentrated adsorbed monovalent:

60 µg of purified VLPs from HPV16 are adsorbed on (50 µg Al³⁺ from Al(OH)₃), at a pH of 6.0 ± 0.2, for one hour at room temperature with gentle stirring. This concentrated adsorbed monovalent is stored at +4°C. Adsorption is checked by centrifuging the preparation and quantifying VLPs in the supernatant.

Preparation of VLP18 concentrated adsorbed monovalent:

60 µg of purified VLPs from HPV18 are adsorbed on (50 µg Al³⁺ from Al(OH)₃), at a pH of 6.0 ± 0.2, for one hour at room temperature with gentle stirring. This concentrated adsorbed monovalent is stored at +4°C. Adsorption is checked by centrifuging the preparation and quantifying VLPs in the supernatant.

3) Final vaccine preparation:

Concentrated adsorbed monovalents prepared by the above method were combined to form a suspension containing 20 µg each VLP per dose. Final vaccine is stored at +4°C.

Addition of VLPs from HPV 31 and 45 at a concentration of 20µg each VLP completes the tetravalent vaccine.

The combined adsorbed bulks, or individual adsorbed bulks, may be further mixed with adjuvants such as 3D-MPL.

Example 2

A Preparation of HPV 16/18 L1 VLPs

Production of HPV 16 and HPV 18 VLPs was carried out using standard protocols as above

B Formation of mixed VLPs

The process of the invention involves disassembly and then reassembly of the HPV 16 and 18 VLPs such that the reassembly of HPV L1 16 and 18 is carried out together to permit the formation of a mixed VLP.

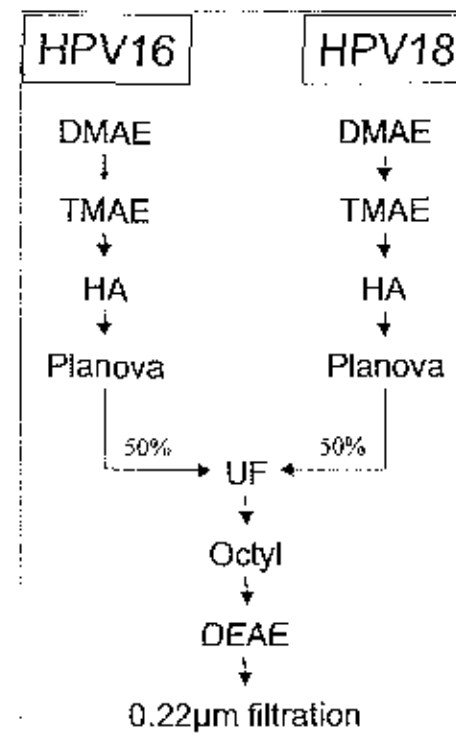
The HPV 16 and 18 VLPs may be combined at any suitable point in the above process prior to the point at which the VLPs are reassembled.

By way of example 2 specific strategies have been tested:

1 mixing of both antigens after the HA step. Based on the L1 concentration in L1A pools, the two components are mixed to reach an equal concentration of HPV16 and 18 to start the UF step. In this case after the ultrafiltration step an Octyl sepharose step is performed as for HPV 18 purification followed by a DEAE step as performed in the HPV 16 procedure.

2 mixing of both extracts and purify. Based on the L1 concentration in Extracts, the two valences are mixed to reach an equal concentration of HPV16 and 18 to start the DMAE step. Again, after the ultrafiltration step an Octyl sepharose step is performed as for HPV 18 followed by a DEAE step as performed in the HPV 16 procedure.

HPV16-HPV18 VLP-mixed at UF step



HPV16-HPV18 VLP-mixed at DMAE step

The same flow sheet is applied but the mixing is performed at the DMAE step instead of the UF step. The concentration used for elution at the anion exchange DMAE step is 200 mM.

Results

HPV16-HPV18 VLP-mixed at UF step

2 lots of mixed VLP's (lot numbers 31B165c and 31B166c) were produced by combining HPV 16 and HPV 18 L1 proteins.

Purity by SDS-Page

The purity of the mixed VLP's was as good as both "classical" HPV 16 or HPV 18 bulks. The purity of the bulks was higher than 95%.

EM data - Fig 1

The EM of 31B165C (UF Retentate after maturation) was compared to a classical HPV16 lot (39B122c). VLP's were well formed, homogeneous in size, without aggregation; some ribbons of capsomeres are present in both experiments.

Size distribution

The size distribution of the VLP's were determined using a Malvern Zetasizer 3000 HS.

The samples were measured undiluted into a plastic cuvette for Malvern analysis (800 µl/cuvette).

The technical conditions were:

- laser wavelength: 532 nm,
- laser power: 50 mW,
- scattered light: detected at 90°.

- temperature: 25°C,
 - duration: automatic determination by the software,
 - number: 3 consecutive measurements,
 - z-average diameter: by cumulants analysis,
- size distribution: by the Contin method.

Classical results for HPV18 L1-VLP's are: 70-80 nm with good polydispersity (<0.1)

Classical results for HPV16 L1-VLP's are: 60-70 nm with good polydispersity (<0.1)

For mixed VLP's, the following results were obtained:

31 B165c : 85 nm with good polydispersity (0.08). VLP's are almost completely formed at the beginning of maturation

31 B166c : 76 nm with good polydispersity (0.08).

The size distribution of lot 31 B165c and 31 B166c as measured by dynamic laser light scattering is shown in figures 2 and 3.

HPV16-HPV18 VLP mixed at DMAE step

Lot n° 31B167B was made up from lots 31B165C05 (HPV18) and 39B167 (HPV16)

Purity by SDS-Page

The purity of the mixed VLP's was as good as both "classical" bulks.

The purity of the bulks was higher than 95%.

Size distribution

The size distribution of the VLP's were determined by using a Malvern Zetasizer 3000 HS.

Classical results for HPV18 L1-VLP's are: 70-80 nm with good polydispersity (<0.1)

Classical results for HPV16 L1-VLP's are: 60-70 nm with good polydispersity (<0.1)

For mixed VLP's, the following results were obtained.

HPV16-HPV18 31B167B : 74 nm with good polydispersity (0.07). VLP's were almost completely formed at the beginning of maturation

Example 3 – Production of a mixed HPV 16, 18, 31, 45 combination vaccine.

Introduction

An immunogenicity study was performed in Balb/C mice using a combination of C-terminally truncated L1 VLPs 16, 18, 31 & 45 adjuvanted with alum + 3D-MPL (herein 'adjuvant A' = 50 µg aluminium salt and 5 µg 3D-MPL)

4 groups of 10 mice were immunised twice intramuscularly on day 0 and 21 respectively with:

1. VLP 31 (2µg) / adjuvant A
2. VLP 45 (2µg) / adjuvant A
3. VLP 16 (2µg) and VLP 18 (2µg) / adjuvant A
4. VLP 16 (2µg), VLP 18 (2µg), VLP 31 (2µg), VLP 45 (2µg) / adjuvant A

Antibody responses against VLPs 16, 18, 31 and 45 were monitored on sera taken at day 35 (14 days post dose II).

Results are shown in Figures 4-7

Antibody response against VLP 16

- Strong antibody responses are induced in post II sera by either VLP16 formulated with VLP 18 on adjuvant A (group 3) or by the full combo (group 4)
- Similar level of antibodies directed against VLP 16 are measured in both groups and no interference is observed.

Antibody response against VLP 18

- Strong antibody responses are induced in post II sera by either VLP18 formulated with VLP 16 on adjuvant A (group 3) or by the full combo (group 4).
- Similar level of antibodies directed against VLP 18 are measured in both groups (less than 1.5 fold difference) and no interference was observed.

Antibody response against VLP 31

- Strong antibody responses are induced in post II sera by either VLP31 formulated alone on adjuvant A (group 1) or by the full combo (group 4).
- Similar level of antibodies directed against VLP 31 are measured in both groups, therefore no interference is observed.

Antibody response against VLP 45

- Strong antibody responses are induced in post II by either VLP 45 formulated alone on adjuvant A (group 2) or by the full combo (group 4).
- Similar level of antibodies directed against VLP 45 are measured in both groups, thus no interference is observed.

Conclusions

No interference is observed when the four VLPs (VLPs 16, 18, 31 & 45) are delivered as a combination.

Claims

- 1 A vaccine composition comprising VLPs containing L1 proteins or functional L1 protein derivatives from HPV 16, HPV 18, HPV 31 and HPV 45 genotypes.
- 2 A vaccine according to claim 1 comprising a mixture of HPV 16 VLPs, HPV 18 VLPs, HPV 31 VLPs and HPV 45 VLPs, each VLP comprising L1 proteins or functional L1 protein derivatives from only 1 HPV genotype.
- 3 A vaccine according to claim 2 wherein the VLPs are L1-only VLPs comprising HPV L1 protein or functional L1 protein derivatives and no other HPV protein.
- 4 A vaccine composition according to any preceding claim comprising VLPs consisting essentially only of proteins from HPV 16, HPV 18, HPV 31 and HPV 45.
- 5 A vaccine composition according to any preceding claim wherein at least one L1 protein is a truncated L1 protein.
- 6 A vaccine composition according to any preceding claim wherein the composition comprises a VLP containing an L1 protein or functional derivative thereof from an additional HPV genotype.
- 7 A vaccine composition according to claim 6 wherein the additional VLP comprises an L1 protein or functional L1 protein derivative selected from one of HPV 33, 52, 53, 58, 35, 56, and 59.
- 8 A vaccine composition according to any preceding claim which is at least 60% effective in preventing cervical cancer.
- 9 A vaccine composition according to any preceding claim additionally comprising an HPV early antigen or immunologically active fragment thereof selected from E1, E2, E3, E4, E5, E6, E7 or E8.

10 A vaccine composition according to any preceding claim formulated with an antigen derived from an organism causing a sexually transmitted disease.

11 A vaccine composition according to claim 10 wherein the antigen is an HSV antigen or immunologically active fragment thereof.

12 A vaccine composition according to claim 10 wherein the antigen is an HIV antigen or immunologically active fragment thereof.

13 A vaccine composition according to claim 10 wherein the antigen is a chlamydia antigen or immunologically active fragment thereof.

14 A vaccine composition according to any preceding claim comprising an HPV L2 protein or fragment thereof.

15 A vaccine composition according to any preceding claim additionally comprising an adjuvant.

16 A vaccine composition according to claim 15 wherein the adjuvant is an aluminium salt.

17 A vaccine composition according to claim 16 wherein the aluminium salt is aluminium hydroxide.

18 A vaccine composition according to claim 16 or 17 additionally comprising 3D-MPL.

19 A vaccine composition according to any of claims 1-18 wherein the VLPs contain L1 proteins which are not chimaeric.

20 A method of vaccine production, the method comprising combining VLPs containing L1 proteins or functional L1 protein derivatives from HPV 16, HPV 18, HPV 31 and HPV 45 to form a vaccine as claimed in any of claims 1-19.

21 Use of a mixture of HPV 16, HPV 18, HPV 31 and HPV 45 VLPs comprising the L1 protein or functional derivative thereof in the preparation of a vaccine for the prevention or treatment of a disorder related to HPV infection.

22 A method of prevention of HPV infection or treatment of disease related to HPV infection, the method comprising delivering to an individual at risk of infection or disease an effective amount of a vaccine composition according to any preceding claim.

23 A stabilised vaccine composition comprising a vaccine according to any of claims 1-18 wherein the VLPs from HPV 16, 18, 31 and 45 are adsorbed onto aluminium hydroxide before mixing.

24 A vaccine composition, method or use according to any of claims 1-23 wherein the immune response against a given VLP type in the vaccine is at least 50 % that of the response of that same VLP type when measured individually.

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SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO
DIRECCIÓN DE NUEVAS CREACIONES

2020
Bogotá, D.C.,

Abogado
ALICIA LLOREDA RICAURTE

ASUNTO	Radicación	06-120701
	Trámite	011
	Evento	379
	Actuación	519
	Oficio	119542
	Folios	012

Patente de Invención	☐	Patente de Modelo de Utilidad	☐
Vía Nacional	☐	Cap. I	☐
Vía PCT (fase nacional)	☐	Cap. II	☐

No. Solicitud Internacional PCT/EP2005/006461
Fecha de Presentación Internacional 14 junio 2005

Como resultado del Examen de Fondo realizado, con fundamento en el Artículo 45 de la Decisión 486 de la Comisión de la Comunidad Andina, se comunica:

1. Documentos estudiados

Descripción:	Folios 105 a 161	29 Noviembre 2006
Reivindicaciones:	Folios 162 a 165	29 Noviembre 2006
Prioridad:	GB 0413510.9	16 Junio 2004
	US 11.114.301	26 Abril 2005

2. Objeto de la invención

Título de la solicitud: "Vacuna contra HPV16 y HPV18 y al menos otro tipo de VPH seleccionado de VPH 31, 45 ó 52"

El problema planteado en esta solicitud consiste en la necesidad de una vacuna que proteja contra los tipos múltiples de VPH (más de 2 clases de VPH).

Para resolver este problema técnico la solicitud en estudio proporciona una composición inmunogénica y una vacuna polivalente que comprende las PVL de VPH 16 y 18 y al menos otro tipo de cáncer de VPH como son VPH 31, 45 ó 52

El objeto de la presente Solicitud se relaciona con la Clasificación Internacional de Patentes (CIP): A61K39/12; A61K39/295; C07K14/025

3. Excepción a la Patentabilidad (Art 20 D 486 literal (d))

El objeto de las reivindicaciones 19 y 21 no es patentable de acuerdo con el Art citado, ya que corresponde a un procedimiento de prevención o tratamiento de

infección de VPH que consiste en administrar una composición o vacuna a un individuo.

4. De la Solicitud de Patente

Reivindicaciones (Art 30 D 486. Art 22 PCT)

- Las reivindicaciones 1 y 20 contienen la expresión "en la que la dosis de la PLV o capsómero de al menos otro tipo de cáncer se reduce con relación al del VPH 16 ó 18" la cual no es clara pues no se establece una relación numérica ni en proporción de los componentes de la composición inmunogénica reclamada, de igual manera esta expresión es relativa y no permite distinguir la invención del estado de la técnica. Se sugiere sustituirla por un término o rango concreto.
- La reivindicación 1 no se encuentra adecuadamente caracterizada puesto que, una composición debe caracterizarse por todos y cada uno de sus componentes y las proporciones de estos dentro de la composición. Se sugiere al solicitante hacer las aclaraciones y modificaciones pertinentes de tal manera que se caracterice adecuadamente el objeto de la invención.
- La reivindicación 8 hace referencia a una composición que contiene VPH 31, VPH 45 y adicionalmente nuevamente hace referencia a VPH 45. Se sugiere hacer la aclaración respectiva.
- No es claro en la reivindicación 9 a que se refieren la expresiones "reivindicaciones preferentes" y "10ug o más" pues no establece la dependencia exacta del objeto de dicha reivindicación y no limita el alcance de la cantidad de componente dentro de la composición. Se sugiere hacer la aclaración respectiva y establecer adecuadamente la dependencia y el rango de concentración del componente o componentes.
- La reivindicación 20 menciona un procedimiento pero este no se define mediante características como alguna etapa ó alguna condición específica en que debiera realizarse. Se sugiere, entonces, definir la invención por sus características esenciales, estructurales o funcionales.
- A lo largo del capítulo reivindicatorio se reclaman diferentes tipos de composiciones que comprenden diversas combinaciones de VHP, proporciones, así como de adyuvantes las cuales llevan a la generación de múltiples posibilidades a reclamar lo cual no define el objeto real a proteger y asimismo, no se encuentran soportadas dentro del capítulo descriptivo. Por lo anterior, se sugiere al solicitante limitar el objeto de la invención.
- En las realizaciones presentadas durante el capítulo descriptivo no se evidencia la elaboración y prueba de composiciones inmunogénicas y/o vacunas que contengan además de las PVL o capsómeros de VPH 16 y VPH 18 otro tipos de cáncer. Por el contrario se establece que solamente con la presencia de estos dos tipos de virus (VPH 16 y VPH 18) se logra una protección cruzada contra otros tipos de cánceres como son 31, y 52 (ver folio 153. líneas 8 a 10, tabla 5). Por lo anterior, el objeto reclamado en la totalidad del capítulo reivindicatorio carece de soporte en la descripción.

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5. Determinación del Estado de la Técnica

El Estado de la Técnica se determinó con base en la fecha de presentación de la solicitud de prioridad (16 Junio 2004) y se encontraron los siguientes documentos que anticipan el objeto de esta solicitud:

Item	Documento	Título	Fecha de Publicación
D1	WO 03/077942	Virus-Like Particles Of Human Papillomavirus	25 Septiembre 2003

D1 (folios 186 a 192) divulga vacunas contra VHP que comprenden partículas semejantes a virus (VPL) de los genotipos de virus VPH 16, 18, 31 y 45.
URL:http://worldwide.espacenet.com/publicationDetails/biblio?DB=EPODOC&I=0&ND=3&adjacent=true&locale=en_gb&FT=D&date=20030925&CC=WO&NR=03077942A2&KC=A2

6. Examen de Patentabilidad (Art 14 D486)

Novedad (Art 16 D486)

La reivindicación 1 consiste en una composición inmunogénica que comprende las PLV o capsómeros de VPH 16 y VPH 18 y al menos otro tipo de cáncer de VPH, el otro tipo de cáncer seleccionándose entre la lista constituida por los tipos de VPH 31, 45 y 52, en la que la dosis de la PLV o capsómero de al menos otro tipo de cáncer se reduce con relación al del VPH 16 ó 18.

Comparación de las características técnicas **esenciales**, con las del Estado de la Técnica:

Características esenciales	D1
Composición inmunogénica	Vacuna tetravalente
PLV o capsómeros de VPH 16 y VPH 18	PLV o capsómeros de VPH 16 y VPH 18
al menos otro tipo de cáncer de VPH (VPH 31, 45 y 52)	PLV o capsómeros de VPH 31 y VPH 45 Y opcionalmente (VPH 52 y VPH 33)

Las características **esenciales** de la composición de la reivindicación 1 habían sido divulgadas previamente en D1, el cual da a conocer composiciones inmunogénicas de tipo vacuna que comprende la combinación de PLVs que contienen proteínas L1 ó derivados funcionales de estas provenientes de genotipos de VPH dentro de los que se encuentran VPH 16, VPH 18, VPH 31 y VPH 45 (Folio 186, pagina 2, línea 9 a 15; folio 187, pagina 6, línea 31 a 33; folio 191, ejemplo 3; reivindicaciones 1 y 4). Dichas combinaciones de pueden darse adicionalmente formando vacunas tetra, penta y hexavalentes adicionando además de los anteriores PLVs otros correspondientes a los genotipos VPH 52, VPH33, entre otros (folio 187, pagina 4, líneas 1 a 5) y sus métodos de elaboración (folio 188, pagina 10, línea 4 a 7; reivindicaciones 20 y 21). Adicionalmente, dichas composiciones o vacunas para su formulación requieren como adyuvantes hidróxido de aluminio, 3DMPL o la mezcla de ambos (folio 188, pagina 9, línea 4 a 12; folio 191, ejemplo 3; reivindicaciones 15 a 18).

Es importante aclarar como se estableció en las objeciones a las reivindicaciones que la expresión "la dosis de la PLV o capsómero de al menos otro tipo de cáncer se reduce con relación al del VPH 16 ó 18", debido a que no define de ninguna manera precisa a que se refiere no puede considerarse como una característica técnica esencial de la composición reclamada.

En consecuencia, la reivindicación 1 carece de novedad, según lo divulgado en el documento D1.

Las reivindicaciones dependientes 2 a 8, 12 a 18 y 20 no mencionan alguna característica que haga nueva a la composición de la reivindicación 1 y que no este presente en el documento D1, por lo cual se considera que no son nuevas.

Nivel Inventivo (Art18 D486)

La **reivindicación 9** consiste en una composición inmunogénica según cualquiera de las reivindicaciones preferentes en la que la composición comprende 10ug, o más, de las PVL de VPH 16 y/o VPH 18 ó capsómero y entre 2-9ug de PLV o capsomero del otro tipo de cáncer.

Comparación de las características técnicas **esenciales**, con las del Estado de la Técnica:

Características esenciales	D1
Composición inmunogénica	Vacuna tetravalente
PLV o capsómeros de VPH 16 y VPH 18 (10ug)	PLV o capsómeros de VPH 16 y VPH 18 (6ug, 10ug, 15ug o 20ug)
otro tipo de cáncer de VPH (2-9ug)	otro tipo de cáncer de VPH (6ug, 10ug, 15ug o 20ug)

El documento D1 se considera el estado de la técnica cercano a la invención definida en la reivindicación 1 porque divulga composiciones inmunogénicas de tipo vacuna que comprende la combinación de PLVs que contienen proteínas L1 ó derivados funcionales de estas provenientes de genotipos de VPH dentro de los que se encuentran VPH 16, VPH 18, VPH 31 y VPH 45 (Folio 186, pagina 2, línea 9 a 15; folio 187, pagina 6, línea 31 a 33; folio 191, ejemplo 3; reivindicaciones 1 y 4). Asimismo revela, que cada vacuna comprende de cada uno de dichos PLVs (VPH 16, VPH 18, VPH 31 y VPH 45) dosis de entre 6 y 20ug que proporcionan una fuerte respuesta inmune contra cada genotipo (folio 188, pagina 10, lineras 1-2). Específicamente establece que la combinación de los cuatro antígenos en cantidades iguales llegan a dicho resultado (folio 191, Ejemplo 3, grupo 4; figuras 4-7)

La diferencia entre la invención, definida en la reivindicación 1 y la composición inmunogénica dada a conocer en D1 consiste en que hay una diferencia en las dosis presentes para cada PLVs presente en la composición y en donde la concentración de los PLVs de VPH 31 y VPH 45 son menores comparados con VPH 16 y VPH 18.

El hecho de modificar las concentraciones o dosis de los PLVs de VPH 31 y VPH 45 disminuyéndolas en comparación con VPH 16 y VPH 18 no se evidencia, puesto que no se presentan realizaciones y/o resultados en donde se evalúe la presencia de los tres o cuatro componentes en una misma composición; por el contrario solo se revela la reacción cruzada usando una vacuna que comprende PLVs de solo dos VPH los cuales corresponde a VPH 16 y VPH 18.

Por lo tanto, el Problema Técnico Objetivo que pretende resolver esta invención se puede formular así: "cómo obtener composiciones inmunogénicas alternativas contra VPH 16 y VPH 18 a las ya conocidas en D1"

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Sin embargo en D1 ya motiva a la persona versada en la materia a modificar las dosis de los PLVs que conforman la vacuna ya que revela que la dosificación de la composición o componentes de la vacuna variarán con el número de tipos de PLV y *que la distribución de dosis debe generar una respuesta inmunológicamente protectora* la cual puede lograrse con dosis entre 6 y 20ug para cada uno de los genotipos de VPH (folio 188, pagina 10, lineas 1-2).

En consecuencia, la persona del oficio normalmente versada en la materia, con la información suministrada en D1 llegaría al objeto de la reivindicación 9 de la solicitud. Por lo cual se considera que no tiene nivel inventivo.

Puesto que las reivindicaciones dependientes 10 y 11 no mencionan características inventivas adicionales tampoco se pueden considerar inventivas.

7. Conclusión

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

Item	Documento	Reivindicaciones afectadas	Requisito que afecta
D1	WO 03/077942	1 a 8, 12 a 18 y 20 9 a 11	Novedad Nivel Inventivo

En cumplimiento del artículo 45 de la Decisión 486 de la Comisión de la Comunidad Andina, se le indica que debe dar respuesta dentro del término de sesenta (60) días contados a partir de la fecha de notificación de la presente comunicación. En caso de no dar respuesta al presente requerimiento, o si a pesar de la respuesta subsistieran los impedimentos para la concesión, se denegará la patente.

Notifíquese el presente oficio conforme a lo dispuesto en el numeral 5.2. literal e), del capítulo quinto del título primero de la Circular Única No. 10 de 2001.



JOSÉ LUIS SALAZAR LÓPEZ
Director de Nuevas Creaciones (C)

El anterior requerimiento fue notificado por fijación en lista, de fecha 10.03.2011

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