Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

Drug Product (DP)

The manufacturing process of the DP is divided into the following critical steps:

- **Preparation of the DS:** (b) (4)
- **Formation of LNP:** In this step, (b) (4)
- **Formulation of the bulk DP:** The bulk DP is formulated by (b) (4)
- **Filling:** The bulk DP is sterile filtered and aseptically filled into 2 mL Type I borosilicate glass vials manufactured by (b) (4) .
- **Labeling and storage:** The filled vials are visually inspected, labeled, and frozen at -90°C to -60°C.

Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

Table 4. Control of COMIRNATY: Tests and Specifications

Quality Attribute	Analytical Procedure	Acceptance Criteria
Appearance	Appearance (Visual)	White to off-white suspension
Appearance (Visible Particulates)	Appearance (Particles) (b) (4)	May contain white to off-white opaque, amorphous particles
(b) (4)	(b) (4)	(b) (4)
(b) (4)	(b) (4)	(b) (4)
(b) (4)	(b) (4)	(b) (4)
LNP (b) (4)	(b) (4)	(b) (4)
LNP (b) (4)	(b) (4)	(b) (4)
RNA (b) (4)	(b) (4) assay	(b) (4)
RNA content	(b) (4) assay	(b) (4)
ALC-0315 content	(b) (4)	(b) (4)
ALC-0159 content	(b) (4)	(b) (4)
DSPC content	(b) (4)	(b) (4)
Cholesterol content	(b) (4)	(b) (4)
Vial content (volume)	Container content	Not less than (b) (4)
Lipid identities	(b) (4)	(b) (4) (ALC-0315, ALC-0159, Cholesterol, DSPC)

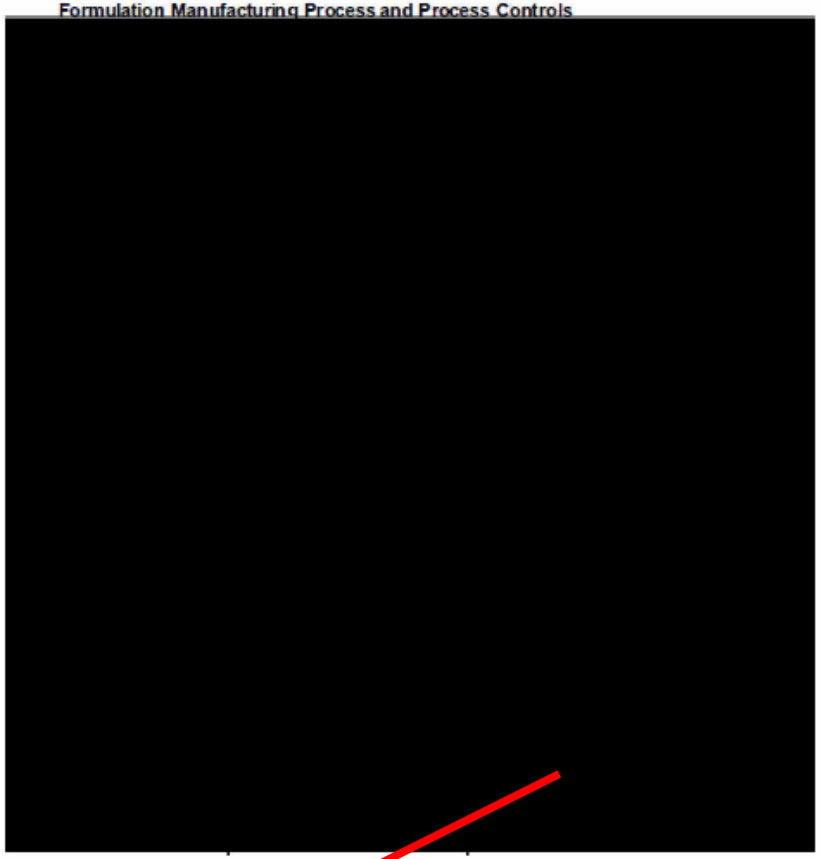
Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

BIONTECH

BioNTech RNA Pharmaceuticals GmbH

Summary Report on Process Performance Qualification for manufacturing of BNT162b2 Bulk DP at mibe and Polymun (R-21-0148)

Figure 1: Flow Diagram of the BNT162b2 LNP Fabrication and Bulk Drug Product Formulation Manufacturing Process and Process Controls



a. [redacted]

Abbreviations: [redacted] ALC-0315 = ((4-hydroxybutyl)azanediyl)bis(hexane-6,1-diyl)bis(2-hexyldecanoate); ALC-0159 = 2-[(polyethylene glycol)-2000]-N,N-ditetradecylacetamide; DSPC = 1,2-distearoyl-sn-glycero-3-phosphocholine; LNP = lipid nanoparticle; IPT-C = in-process test for control; [redacted]; PBS = phosphate-buffered saline; $Na_2HPO_4 \cdot 2\ H_2O$ = dibasic sodium phosphate, dihydrate; KCl = potassium chloride; KH_2PO_4 = monobasic potassium phosphate; NaCl = sodium chloride

Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

BIONTECH

BioNTech RNA Pharmaceuticals GmbH

Summary Report on Process Performance Qualification for manufacturing of BNT162b2 Bulk DP at mibe and Polymun (R-21-0148)

Attachment 1: Critical Process Parameters – Bulk DP Process

CPPs (italic = non CPP)	Operating range	Polymun (Phase I PPQ)	Polymun (Phase II PPQs)			Mibe		
		BCV4/L15	BCV4/L19	BCV4/L20	BCV4/L21	201113	210101	210103
[redacted]								
[redacted]						N/A	N/A	N/A
[redacted]						N/A	N/A	N/A

Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

BIONTECH

BioNTech RNA Pharmaceuticals GmbH

Summary Report on Process Performance Qualification for manufacturing of BNT162b2 Bulk DP at mibe and Polymun (R-21-0148)

Parameter (Italic: non critical)	Acceptance criteria	Polymun (Phase I PPQ)	Polymun (Phase II PPQs)			Mibe (Phase I PPQ)	Mibe (Phase II PPQs)	
		BCV4/L15	BCV4/L19	BCV4/L20	BCV4/L21	201113	210101	210103
Aqueous Phase Preparation								
RNA content (mg/mL)	[redacted]							
LNP Preparation								
RNA content (mg/mL)	[redacted]							
Particle size (nm)	[redacted]							
Polydispersity index	[redacted]							
TFF								
Appearance	[redacted]							
RNA content (mg/mL)	[redacted]							
RNA encapsulation (%)	[redacted]							
ALC-0315 content (mg/mL)	[redacted]							
ALC-0159 content (mg/mL)	[redacted]							
DSPC content (mg/mL)	[redacted]							
Cholesterol content (mg/mL)	[redacted]							

Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

1.2. Analytical Comparability Testing Strategy

The panel of tests, both release and heightened characterization, performed to evaluate drug product comparability are shown in Table 2. Additional heightened characterization methods were performed to evaluate selected drug product LNP and purity attributes.

Table 2. BNT162b2 Drug Product Comparability Testing Panel

Quality Attribute	Analytical Procedure	Release / Characterization
Composition and Strength		
Appearance	Appearance (Visual)	Release
Appearance (Visible Particulates)	Appearance (Particles)[a]	Release
Subvisible Particles	Subvisible Particulate Matter[a, b]	Release
pH	Potentiometry[a]	Release
Osmolality	Osmometry[a, c]	Release
LNP Size	Dynamic Light Scattering (DLS)	Release
LNP Polydispersity	Dynamic Light Scattering (DLS)	Release
RNA Encapsulation	Fluorescence assay	Release
RNA content	Fluorescence assay	Release
ALC-0315 content	HPLC-CAD	Release
ALC-0159 content	HPLC-CAD	Release
DSPC content	HPLC-CAD	Release
Cholesterol content	HPLC-CAD	Release
Surface Charge	[redacted]	Characterization
Size Distribution and Shape		Characterization
Surface PEG Characterization		Characterization
Identity		
Lipid identities	HPLC-CAD	Release
Identity of encoded RNA sequence	RT-PCR	Release
Potency		
In Vitro Expression	Cell-based Flow Cytometry	Release
Purity		
RNA Integrity	Capillary Gel Electrophoresis	Release
5'- Cap	RP-HPLC[d]	Characterization
Poly(A) Tail	ddPCR	Characterization
Poly(A) Tail: Length and Distribution	RP-HPLC[d]	Characterization

a. Compendial
b. USP<787> (obscuration method), and aligned with upcoming (Jan 2021) revision of Ph. Eur. 2.9.19
c. USP<785>; also in accordance with Ph Eur. 2.2.35, with minor difference in instrument calibration
d. Tested side-by-side

Abbreviations: LNP = Lipid nanoparticles; CAD = charged aerosol detector; RT-PCR = reverse transcription polymerase chain reaction; [redacted] ddPCR = droplet digital PCR; qPCR = quantitative PCR

Australian Government
Department of Health
Therapeutic Goods Administration

Nonclinical Evaluation Report

BNT162b2 [mRNA] COVID-19 vaccine (COMIRNATY™)

Submission No: PM-2020-05461-1-2
Sponsor: Pfizer Australia Pty Ltd

January 2021

Page 46 of 58

4.3. METABOLISM

In vitro and *in vivo* studies were conducted to evaluate metabolism of novel lipids ALC-0159 and ALC-0315. The metabolism studies used LC/MS-MS bioanalytical methods for the detection of novel lipids and their metabolites. The major metabolic pathways for ALC-0159 and ALC-0315 in various species, as proposed by the sponsor, are shown in Figure 4-7 and Figure 4-8, respectively.

[redacted]

[redacted]

In vitro studies indicated minor metabolism of ALC-0159 and ALC-0315 in liver, with most of the lipids found unchanged at the end of the 2 or 4 h incubation period (Table 4-3 and Table 4-4).

Vaccino Pfizer/BioNTech «Comirnaty Covid-19 mRNA BNT162b2»

COMPOSIZIONE QUALI-QUANTITATIVA

Ingrediente	Funzione	Quantità per dose
BNT162b2	Attivo	30 µg
ALC-0315	Lipide funzionale	0,43 mg
ALC-0159	Lipide funzionale	0,0534 mg
DSPC	Lipide strutturale	0,09 mg
Colesterolo	Lipide strutturale	0,2 mg
Saccarosio	Crioprotettivo	6 mg
Cloruro di sodio	Elettrolita: componente tampone pH	2,52 mg
Cloruro di potassio	Elettrolita: componente tampone pH	0,01 mg
Fosfato disodico diidrato	Elettrolita: componente tampone pH	0,07 mg
Diidrogenofosfato di potassio	Elettrolita: componente tampone pH	0,01 mg
Acqua per inoculazioni	Mezzo disperdente	q.s. a 0.3 ml

TESTO ATTO

Atto Camera

Interrogazione a risposta scritta 4-12281

presentato da

CUNIAL Sara

testo di

13 Giugno 2022

Lunedì 13 giugno 2022, seduta n. 706

CUNIAL. — Al Ministro della salute. — Per sapere – premesso che:

con l'interrogazione n. 4-08164, l'interrogante si era già occupata della problematica dei due nuovi eccipienti mai autorizzati prima nel vaccino COVID-19 di Pfizer: il lipide cationico ALC-0315 e il lipide PEGilato ALC-0159;

con l'articolo scientifico dal titolo «Criticità chimico-fisiche e potenziale tossicologico dei nanomateriali lipidici contenuti nel vaccino Comirnaty/Pfizer» il ricercatore biochimico indipendente Gabriele Segalla esamina alcune evidenti criticità chimico-fisiche del preparato, in merito alla manifesta instabilità della sua composizione quali-quantitativa, nonché al suo conseguente potenziale tossicologico, nella fattispecie correlato alla possibile formazione di ROS (reactive oxygen species), dopo l'inoculazione intramuscolare, in diversi siti biologici, quali, potenzialmente, reni, fegato, cuore, cervello, e altro, provocandone disfunzioni e alterazioni;

lo studioso conclude affermando che il vaccino Comirnaty Covid-19 mRNA BNT162b2, nella sua versione e composizione originale, denominata PBS/Sucrose, presenta numerose criticità e non conformità, esaminate nel dettaglio nello studio e qui appresso sinteticamente elencate:

i due eccipienti funzionali preposti alla formazione di nanoparticelle lipidiche, ALC-0315 e ALC-0159, non sono registrati in nessuna farmacopea, né rientrano tra le sostanze esaminate e classificate in conformità al regolamento (CE) CLP n. 1272/2008 relativo alla classificazione, all'etichettatura e all'imballaggio delle sostanze e delle miscele in Europa;

tali eccipienti non compaiono neppure nell'inventario delle sostanze registrate in conformità al regolamento (CE) REACH n. 1907/2006 concernente la registrazione, la valutazione, l'autorizzazione e la restrizione delle sostanze chimiche in Europa. Non se ne conosce pertanto il profilo tossicologico in primis;

non sono state espletate tutte le procedure di analisi chimico-fisica e test tossicologici previste per le nanoforme di dette sostanze, in contrasto con quanto dettato dal regolamento (UE) 2018/1881 che modifica il regolamento (CE) n. 1907/2006 del Parlamento europeo e del Consiglio concernente la registrazione, la valutazione, l'autorizzazione e la restrizione delle sostanze chimiche (REACH), per ricomprendervi le nanoforme delle sostanze;

non sono stati effettuati, con il consenso dell'ente certificatore, studi di cancerogenicità, genotossicità e mutagenicità del preparato, nonostante sia stato ormai comprovato da numerosi studi scientifici come le Specie reattive dell'ossigeno (ROS) generate dalle nanoparticelle possano avere un alto potenziale cancerogeno, genotossico e mutageno;

INTERROGAZIONE A RISPOSTA SCRITTA 4/12281 CAMERA

TESTO ATTO

Atto Camera

Interrogazione a risposta scritta 4-12281

…con l'articolo scientifico dal titolo «Criticità chimico-fisiche e potenziale tossicologico dei nanomateriali lipidici contenuti nel vaccino Comirnaty/Pfizer» il ricercatore biochimico indipendente Gabriele Segalla esamina alcune evidenti criticità chimico-fisiche del preparato, in merito alla manifesta instabilità della sua composizione quali-quantitativa, nonché al suo conseguente potenziale tossicologico, nella fattispecie correlato alla possibile formazione di ROS (reactive oxygen species), dopo l'inoculazione intramuscolare, in diversi siti biologici, quali, potenzialmente, reni, fegato, cuore, cervello, e altro, provocandone disfunzioni e alterazioni…

DISINFECTION
Quaderno monografico 02/2022

Ottobre 2022

www.ndmagazine.it/

GABRIELE SEGALLA

Criticità chimico-fisiche e potenziale tossicologico dei nanomateriali lipidici contenuti in un vaccino a mRNA

DIFFUSIONE RISERVATA AGLI OPERATORI SANITARI

Criticità chimico-fisiche e potenziale tossicologico dei nanomateriali lipidici contenuti in un vaccino a mRNA

Gabriele Segalla
gabriele.segalla@gmail.com
Biochimico
Ricercatore indipendente *

SOMMARIO

Il preparato medicinale denominato Comirnaty di Pfizer-BioNTech è una sospensione acquosa di nanomateriali lipidici, destinata a costituire, dopo le fasi di scongelamento e diluizione, il prodotto finito iniettabile per via intramuscolare.

Nel presente studio, si esaminano alcune evidenti criticità chimico-fisiche del preparato, in merito alla manifesta instabilità della sua composizione quali-quantitativa, nonché al suo conseguente potenziale tossicologico, nella fattispecie correlato alla possibile formazione di ROS (*reactive oxygen species*), dopo l'inoculazione intramuscolare, in diversi siti biologici, quali, potenzialmente, reni, fegato, cuore, cervello, ecc., provocandone disfunzioni e alterazioni.

Particolare preoccupazione desta la presenza nel formulato dei due eccipienti funzionali, ALC-0315 e ALC-0159, mai usati prima in un prodotto medicinale, né registrati in Farmacopea Europea, né tantomeno nell'inventario europeo C&L.

Le attuali Schede di Dati di Sicurezza del fabbricante risultano omissive e inadempienti, soprattutto in merito a quanto previsto dalle vigenti normative europee in tema di registrazione, valutazione, autorizzazione e restrizione dei nanomateriali.

La presenza di elettroliti nel preparato e la successiva fase di diluizione dopo lo scongelamento e prima dell'inoculazione destano fondate preoccupazioni sulla precaria stabilità della sospensione risultante e sull'Indice di polidispersione dei nanomateriali in essa contenuti, fattori questi ipotizzabili come le *root causes* di numerosi effetti avversi post-vaccinazione registrati a livello statistico-epidemiologico.

Si consigliano e auspicano ulteriori immediati studi e verifiche in merito, prendendo nel frattempo in considerazione, se del caso e a scopo meramente precauzionale, l'immediata sospensione delle vaccinazioni con il preparato Comirnaty di Pfizer-BioNTech, alla luce dell'articolo 10 del Codice di Norimberga: *"Durante l'esperimento lo scienziato responsabile deve essere pronto a interromperlo in qualunque momento se indotto a credere che la continuazione dell'esperimento comporterebbe probabilmente lesioni, invalidità o morte per il soggetto umano"*.

*** Dr. Gabriele Segalla,**
Chimico, Specialista in Chimica organico-biologica, Chimica cosmetica e delle preparazioni ad uso topico per medicina estetica, Chimica delle micro-emulsioni e dei sistemi colloidali, Chimica delle sostanze organiche naturali.

E-mail: gabriele.segalla@gmail.com

L'Autore dichiara di avere eseguito la ricerca riportata nel presente lavoro in qualità di Ricercatore Indipendente, di non aver ricevuto alcun finanziamento e di non aver conflitti di interesse.

International Journal of Vaccine Theory, Practice, and Research

IJVTPR

Chemical-physical criticality and toxicological potential of lipid nanomaterials contained in a COVID-19 mRNA vaccine

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Multichem R&D

ORCID: https://orcid.org/0000-0002-5969-3732

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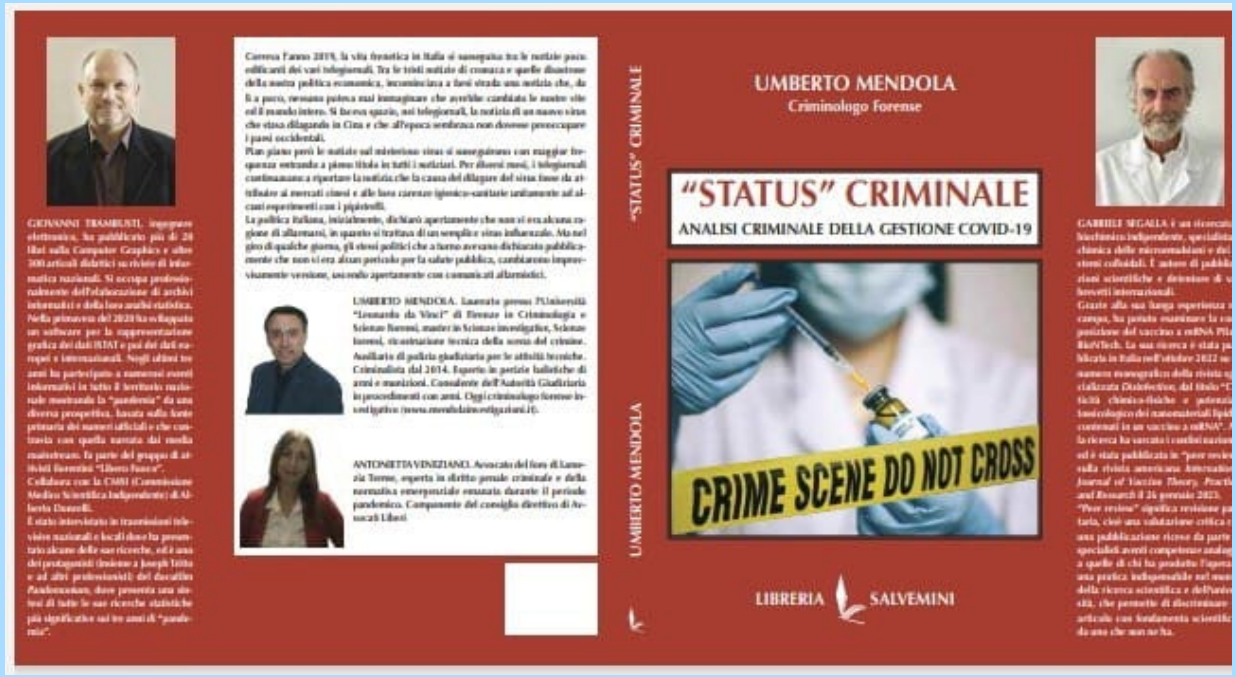
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ANALISI CRIMINALE DELLA GESTIONE COVID-19
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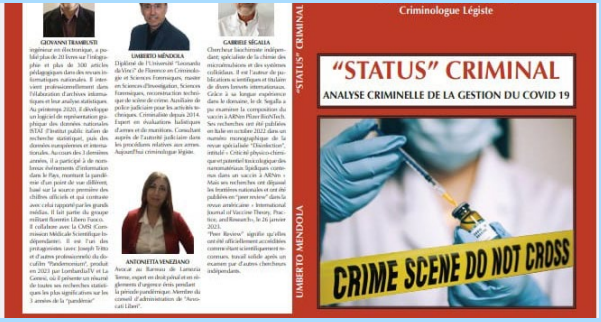
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КРИМИНАЛЬНЫЙ СТАТ
СУДЕБНЫЙ КРИМИНАЛИСТ
КРИМИНАЛЬНЫЙ СТАТУС
Криминалистическая экспертиза борьбы с ковидом
Умберто Мендола
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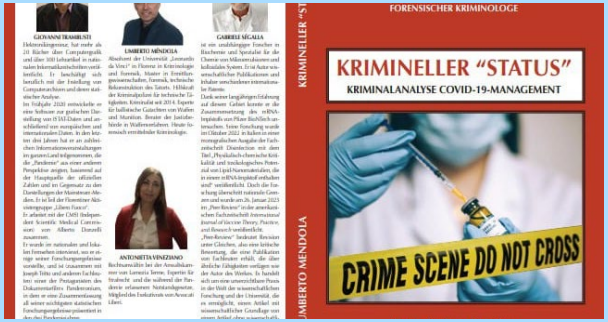
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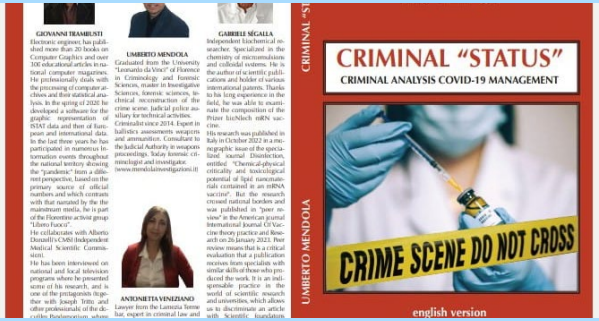
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International Journal of Vaccine Theory, Practice, and Research

Apparent Cytotoxicity and Intrinsic Cytotoxicity of Lipid Nanomaterials Contained in a COVID-19 mRNA Vaccine

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EUROPEAN
COMMISSION

22 November 2017

Guidelines on Good Manufacturing Practice specific to Advanced Therapy Medicinal Products

14.16. Quality defect records should be retained and used to evaluate the possible existence of recurring problems. Competent authorities should be informed in a timely manner in case of a confirmed quality defect (faulty manufacture, product deterioration, detection of falsification, non-compliance with the marketing authorisation or Product Specification File, or any other serious quality problems) with an ATMP which may result in the recall of the product or an abnormal restriction in the supply. Unplanned deviations as described in Section 11.4 should not be notified.

EUROPEAN COMMISSION

22 November 2017

Guidelines on Good Manufacturing Practice specific to Advanced Therapy Medicinal Products

DIFETTI DI QUALITÀ (QUALITY DEFECTS):
DIFETTI LEGATI A ERRORI RELATIVI ALLA COMPOSIZIONE, ALLA FABBRICAZIONE, ALLA STABILITÀ, ALLA FALSIFICAZIONE E NON CONFORMITÀ CON QUANTO SANCITO DALL'AUTORIZZAZIONE ALL'IMMISSIONE IN COMMERCIO, ECC.

GRAVI DIFETTI DI QUALITÀ
(CRITICAL QUALITY DEFECTS):
DIFETTI, DOVUTI AD UN MANCATO RISPETTO DELLE BUONE PRASSI DI FABBRICAZIONE DELL'UE, E POTENZIALMENTE PERICOLOSI PER LA VITA O CHE POTREBBERO CAUSARE UN GRAVE RISCHIO PER LA SALUTE

International Journal of Vaccine Theory, Practice, and Research

Adjuvant Activity and Toxicological Risks of Lipid Nanoparticles Contained in the COVID-19 "mRNA Vaccines"

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WHO Technical Report Series, No. 927, 2005

Annex 1

WHO guidelines on nonclinical evaluation of vaccines

Adjuvants

Substances that are intended to enhance relevant immune response and subsequent clinical efficacy of the vaccine.

ADIUVANTI: sostanze che hanno lo scopo di aumentare la risposta immunitaria in modo rilevante

WHO Expert Committee on Biological Standardization

Sixty-fourth report

Annex 2

Guidelines on the nonclinical evaluation of vaccine adjuvants and adjuvanted vaccines

Novel adjuvant: a novel adjuvant is an adjuvant that has not been contained in a licensed vaccine.

NUOVO ADIUVANTE è un adiuvante che non è mai stato utilizzato prima in un vaccino autorizzato

WHO Technical Report Series, No. 927, 2005

Annex 1

WHO guidelines on nonclinical evaluation of vaccines

4.2.3 *Genotoxicity and carcinogenicity studies*

Genotoxicity studies are normally not needed for the final vaccine formulation. However, they may be required for particular vaccine components such as novel adjuvants and additives. If needed, the in vitro tests for mutations and chromosomal damage should be done prior to first human exposure. The full battery of tests for genotoxicity may be performed in parallel with clinical trials (*28*).

Carcinogenicity studies are not required for vaccine antigens. However, they may be required for particular vaccine components such as novel adjuvants and additives.

WHO Technical Report Series, No. 927, 2005

Annex 1

WHO guidelines on nonclinical evaluation of vaccines

4.2.3 *Genotoxicity and carcinogenicity studies*

Di norma non sono necessari studi di genotossicità per la formulazione finale del vaccino. Tuttavia, possono essere richiesti per particolari componenti del vaccino, come nuovi adiuvanti e additivi…

WHO Technical Report Series, No. 927, 2005

Annex 1

WHO guidelines on nonclinical evaluation of vaccines

4.2.3 *Genotoxicity and carcinogenicity studies*

Gli studi di cancerogenicità non sono necessari per gli antigeni dei vaccini. Tuttavia, possono essere richiesti per particolari componenti del vaccino, come nuovi adiuvanti e additivi.

EPAR

European public assessment report

COMIRNATY, COMIRNATY ORIGINAL/OMICRON BA.1, COMIRNATY ORIGINAL/OMICRON BA.4-5, COMIRNATY OMICRON XBB.1.5
(COVID-19 mRNA VACCINE)
RISK MANAGEMENT PLAN

OCTOBER 2023

a. Safety pharmacology, genotoxicity, and carcinogenicity studies were not conducted, in accordance with 2005 WHO vaccine guideline, as they are generally not considered necessary to support development and licensure of vaccines for infectious diseases. In addition, the components of the vaccine construct are lipids and RNA and are not expected to have carcinogenic or genotoxic potential.

p. 65

EPAR

European public assessment report

COMIRNATY, COMIRNATY ORIGINAL/OMICRON BA.1, COMIRNATY ORIGINAL/OMICRON BA.4-5, COMIRNATY OMICRON XBB.1.5
(COVID-19 mRNA VACCINE)
RISK MANAGEMENT PLAN

OCTOBER 2023

Non sono stati condotti studi di sicurezza farmacologica, genotossicità e cancerogenicità, in conformità con le linee guida sui vaccini dell'OMS del 2005, in quanto generalmente non sono considerati necessari per sostenere lo sviluppo e l'autorizzazione di vaccini per le malattie infettive. Inoltre, i componenti del costrutto vaccinale sono lipidi e RNA e non si prevede che abbiano un potenziale cancerogeno o genotossico.

COMIRNATY, COMIRNATY ORIGINAL/OMICRON BA.1, COMIRNATY ORIGINAL/OMICRON BA.4-5, COMIRNATY OMICRON XBB.1.5 (COVID-19 mRNA VACCINE) RISK MANAGEMENT PLAN

Module SVII. Identified and Potential Risks

- **The vaccine construct and the formulation**. The COVID-19 mRNA vaccine consists of non-infectious, non-replicating RNA in a lipid-based formulation, which delivers the RNA to cells in the immunised person. Protein expression from the RNA is transient, and as is RNA itself. There is no systemic toxicity associated with the LNP or its metabolism (Study reports 38166 and 20GR142). Vacuolation of hepatocytes was observed in rat toxicity studies and believed to be associated with the uptake of the LNP and was without evidence of any effect on liver function. The liver vacuolation was reversed approximately 3-weeks after the last administration.

- **The degradation of the active substance / antigen and potential impact on safety related to this; (e.g., for mRNA-based vaccines).** Like endogenous mRNA in the cytosol, vaccine RNA in cytosol is degraded. The COVID-19 mRNA contains no known toxic products of the degradation of the RNA or the lipids in the formulation.

- **The vaccine does not contain an adjuvant.**

EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

Assessment report
Comirnaty

Common name: COVID-19 mRNA vaccine (nucleoside-modified)
Procedure No. EMEA/H/C/005735/0000
19 February 2021
EMA/707383/2020 Corr.1
Committee for Medicinal Products for Human Use (CHMP)

Toxicology Page 55/140

With regards to the vaccine components, only the whole formulation (modified RNA in LNPs) were used, so there is no toxicological data on the LNP alone or its specific novel excipients. The novel LNP components, these are not considered primarily as adjuvant substances.

No genotoxicity nor carcinogenicity studies have been provided. The components of the vaccine formulation are lipids and RNA that are not expected to have genotoxic potential.

I nuovi componenti costituiti dalle LNP non sono considerati primariamente come sostanze adiuvanti

Nanovaccines and their mode of action

Mehfuz Zaman [a], Michael F. Good [b], Istvan Toth [a c]

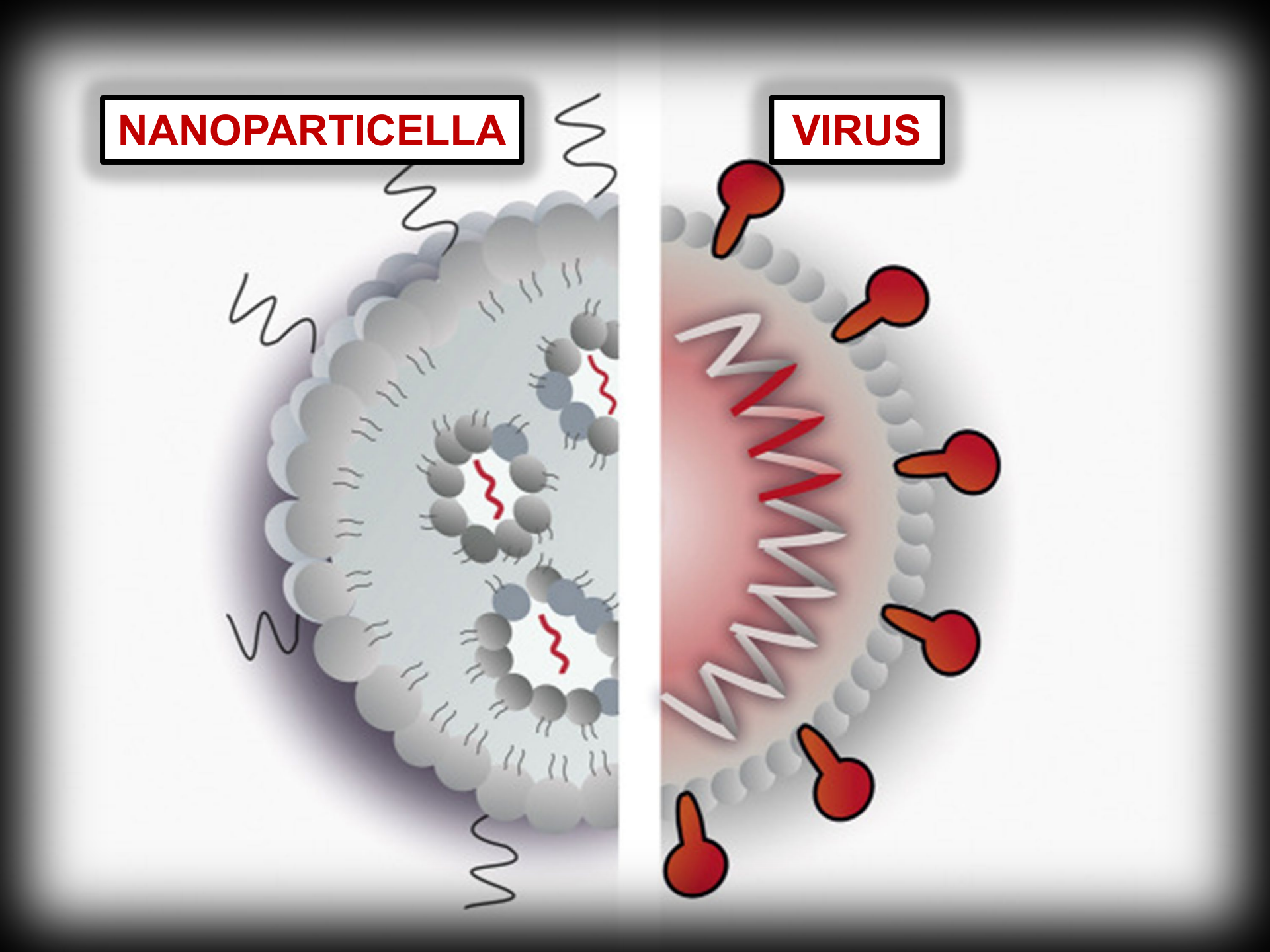
Methods

Nanoparticle adjuvanticity

Volume 60, Issue 3, 1 May 2013, Pages 226-231

Adjuvants act to stimulate and augment immune responses to antigens. An extensive number of studies have reported nanoparticles as adjuvants. In many of these studies, nanoparticles induced similar or higher immune responses than aluminum-containing adjuvants (most widely used in human) such as alum [10], [23]. Also, cobalt oxide nanoparticles stimulated less allergic antibody production and in-vivo inflammation at different administration sites than an alum based adjuvant [23]. Therefore the...

Nanovaccines are currently an area receiving a high level of interest. Interactions between nanovaccines and cells of the immune system generally depend on particle characteristics such as size and surface properties including surface charge and hydrophobicity. Consequently these parameters can influence the uptake of nanovaccines, their antigenicity, adjuvant activity, and inflammatory response. Nanotechnology is actively being investigated to design vaccine carriers, adjuvants, and drug...


NANOPARTICELLA
VIRUS

Article

COVID-19 vaccine BNT162b1 elicits human antibody and T_H1 T cell responses

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Check for updates

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Lipid nanoparticle (LNP)-formulated mRNA vaccine technology allows the delivery of precise genetic information together with an adjuvant effect to antigen-presenting cells[4]. The prophylactic effectiveness of this technology against multiple viral targets has been proven in preclinical models[5–7]. LNP- and liposome-formulated RNA

ARTICLES
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nature immunology

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IL-1 and IL-1ra are key regulators of the inflammatory response to RNA vaccines

Siri Tahtinen[1], Ann-Jay Tong[1], Patricia Himmels[1], Jaehak Oh[1], Andres Paler-Martinez[1], Leesun Kim[1], Sara Wichner[1], Yoko Oei[1], Mark J. McCarron[1], Emily C. Freund[1], Zhainib Adel Amir[1], Cecile C. de la Cruz[1], Benjamin Haley[1], Craig Blanchette[1], Jill M. Schartner[1], Weilan Ye[1], Mahesh Yadav[1], Ugur Sahin[2], Lélia Delamarre[1] and Ira Mellman[1]✉

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Competing interests

All authors except U.S. are current or former employees of Genentech (South San Francisco, USA). U.S. is a management board member and an employee at BioNTech AG (Mainz, Germany), and inventor on patents and patent applications related to RNA vaccines used in this study. U.S. has securities from BioNTech AG.

Immunity

Lipid nanoparticles enhance the efficacy of mRNA and protein subunit vaccines by inducing robust T follicular helper cell and humoral responses

Alameh et al., 2021, Immunity 54, 2877–2892
December 14, 2021 © 2021 Elsevier Inc.
https://doi.org/10.1016/j.immuni.2021.11.001

Mohamad-Gabriel Alameh,[1,10] István Tombácz,[1,10] Emily Bettini,[2,10] Katlyn Lederer,[2] Sonia Ndeupen,[3] Chutamath Sittplangkoon,[4] Joel R. Wilmore,[5] Brian T. Gaudette,[5] Ousamah Y. Soliman,[1] Matthew Pine,[1] Philip Hicks,[2,6] Tomaz B. Manzoni,[2] James J. Knox,[5] John L. Johnson,[5] Dorottya Laczkó,[1] Hiromi Muramatsu,[1] Benjamin Davis,[1] Wenzhao Meng,[5] Aaron M. Rosenfeld,[5] Shirin Strohmeier,[7] Paulo J.C. Lin,[8] Barbara L. Mui,[8] Ying K. Tam,[8] Katalin Karikó,[1,9] Alain Jacquet,[4] Florian Krammer,[7] Paul Bates,[2] Michael P. Cancro,[5] Drew Weissman,[1] Eline T. Luning Prak,[5] David Allman,[5] Botond Z. Igyártó,[3] Michela Locci,[2,*] and Norbert Pardi[1,11,*]

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[8]Acuitas Therapeutics, Vancouver, BC, Canada
[9]BioNTech RNA Pharmaceuticals, Mainz, Germany

KATALIN KARIKO' - DREW WEISSMAN
PREMIO NOBEL PER LA MEDICINA 2023

Immunity

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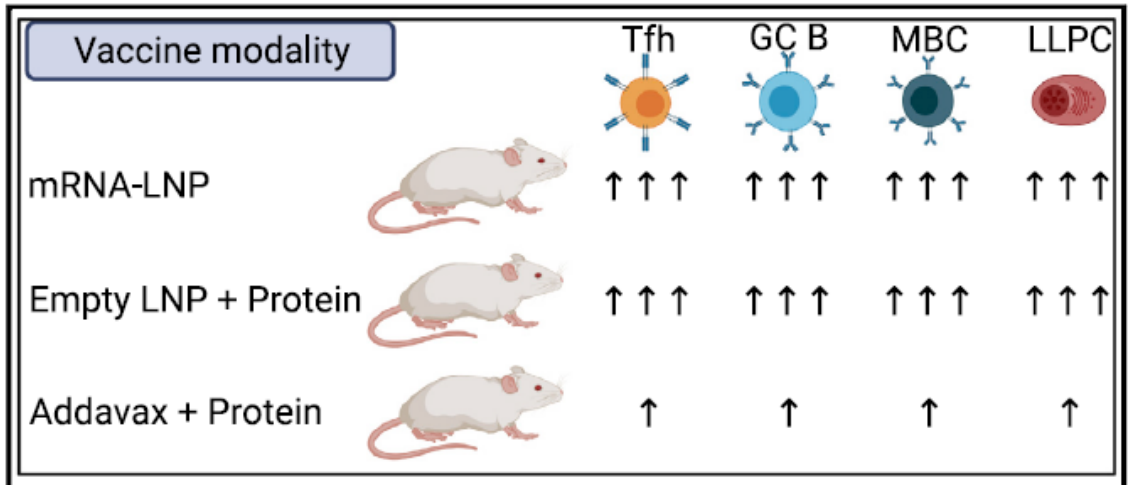

The mechanism of action of nucleoside-modified mRNA-LNP vaccines is unknown. Alameh et al. demonstrate that LNPs can possess adjuvant activity and promote robust induction of Tfh cell, B cell, and humoral responses when utilized in mRNA and protein subunit vaccines in mice. IL-6 induction and the ionizable lipid component are critical for the adjuvant activity of LNPs.

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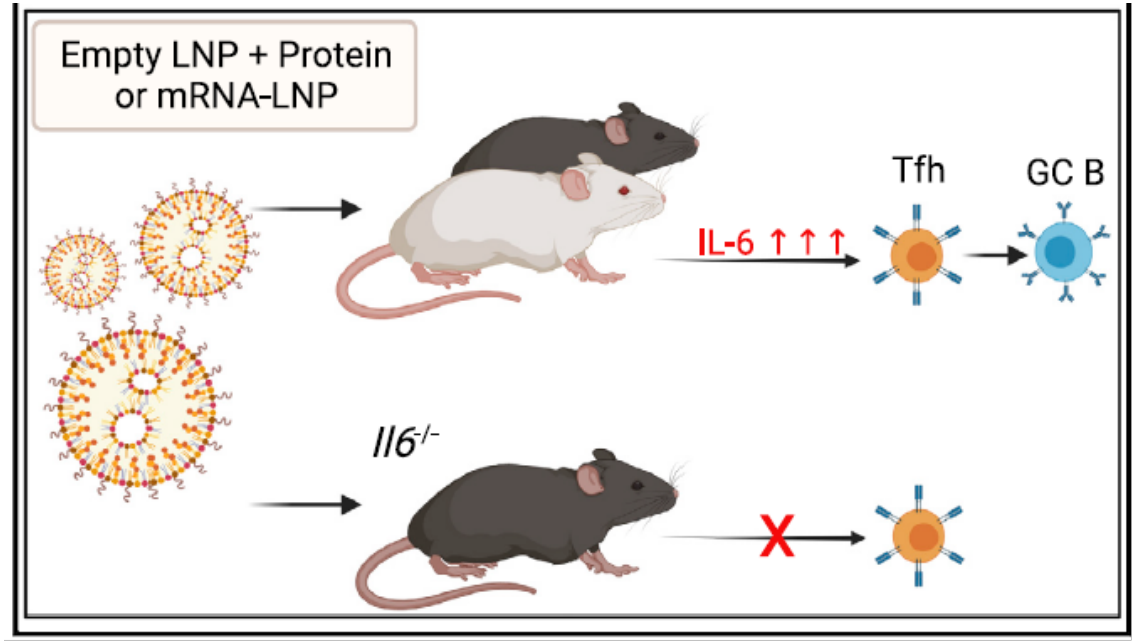

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Innate immune mechanisms of mRNA vaccines

Rein Verbeke,[1,2,*] Michael J. Hogan,[3] Karin Loré,[4,5] and Norbert Pardi[6,*]

IONIZABLE LIPID NANOPARTICLES PROVIDE POTENT ADJUVANT ACTIVITY

Immunity 55, November 8, 2022

iLNPs function as more than delivery agents. In fact, both empty iLNPs and mRNA-iLNP complexes serve as extremely effective adjuvants. Empty iLNPs were successfully used to adjuvant vaccines containing hepatitis B virus or dengue virus protein antigens (Swaminathan et al., 2016a, 2016b), and modified mRNA-iLNPs encoding luciferase were used to strongly adjuvant an influenza HA protein vaccine (Pardi et al., 2018). More recently, empty iLNPs mixed with influenza HA or SARS-CoV-2 spike receptor binding domain (RBD) proteins were shown to induce much stronger immune responses in mice than AddaVax-adjuvanted protein vaccines (Alameh et al., 2021). Of note, iLNP-ad-

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SCIENCE MEDICINES HEALTH

Quality rolling review CHMP overview and list of questions

COVID-19 mRNA Vaccine BioNTech

EMA QUALITY ROLLING REVIEW – CHMP - AMSTERDAM, **30 NOVEMBER 2020**
EMA/CHMP/641856/2020

Results from the Luminex-based [redacted] assay revealed that immunisation with LNP formulated luciferase modRNA transiently increased levels of MCP-1, IL-6, IP-10, at 6 hours post-immunization. These results in mice suggest that the LNP can activate the innate immune system of mice, by the synthesis of pro-inflammatory cytokines. As the effect was transient, this effect could be considered as an adjuvant-like effect. Overall, on the basis of above, the LNP formulation is expected to have not only a role to protect modRNA from nucleases degradation, and facilitating cellular transfection, but also adjuvant like effects.

In view of potential acute immunotoxicity mediated by LNPs, does the Applicant possess data on other timepoints (earlier than 6h or beyond) regarding the cytokines measurements? **(OC)**

The Applicant is also asked to discuss the absence of an in vitro hPBMC stimulation assay in healthy donors to assess reactogenicity **(OC)**.

Extrapolating to clinics, the Applicant is requested to discuss the level of IL-6 cytokine induced by LNPs, considering that asymptomatic but infected subjects candidate to vaccination, could display higher IL-6 levels during early phase infection **(OC)**.

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Page 52/79

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… Nel complesso, sulla base di quanto sopra, ci si aspetta che la formulazione a base di LNP abbia non solo un ruolo nel proteggere l'RNA dalla degradazione delle nucleasi e nel facilitare la trasfezione cellulare, ma anche effetti di tipo adiuvante.

EPAR

European public assessment report

COMIRNATY, COMIRNATY ORIGINAL/OMICRON BA.1,
CO[illegible]ON

IL VACCINO NON CONTIENE UN ADIUVANTE

- The vaccine does not contain an adjuvant.

p. 112

EMA QUALITY ROLLING REVIEW – CHMP
COVID-19 mRNA Vaccine Pfizer BioNTech
30 NOVEMBER 2020

EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

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LNP = ADIUVANTI

EMA EPAR – RISK MANAGEMENT PLAN
COVID-19 mRNA Vaccine Pfizer BioNTech
OCTOBER 2023

EPAR
European public assessment report

COMIRNATY, COMIRNATY ORIGINAL/OMICRON BA.1, COMIRNATY ORIGINAL/OMICRON BA.4-5, COMIRNATY OMICRON XBB.1.5
(COVID-19 mRNA VACCINE)
RISK MANAGEMENT PLAN OCTOBER 2023

a. Safety pharmacology, genotoxicity, and carcinogenicity studies were not conducted, in accordance with 2005 WHO vaccine guideline, as they are generally not considered necessary to support development and licensure of vaccines for infectious diseases. In addition, the components of the vaccine construct are lipids and RNA and are not expected to have carcinogenic or genotoxic potential.

p. 65

- **The vaccine does not contain an adjuvant.**

p. 112

LNP = NON ADIUVANTI

La «FRAUD-Chart»

EPAR/RMP di Pfizer/BioNTech afferma: Test di **Genotossicità** & **Cancerogenicità NON** necessari. Il Vaccino **NON** contiene un **ADIUVANTE**

LNP sono da considerarsi ADIUVANTI ?

SI

Il Vaccino **CONTIENE ADIUVANTI**. **EPAR/RMP di Pfizer/BioNTech FALSO** OMISSIONE DOLOSA DI TEST di **Genotossicità** & **Cancerogenicità**

FRAUD

PRODOTTO NON TESTATO, PERICOLOSO PER LA SALUTE UMANA. AUTORIZZAZIONE ALL'IMMISSIONE IN COMMERCIO INVALIDA

NO

Precedenti STUDI di Pfizer/BioNTech che affermano: LNP = ADIUVANTI

FAKE

1. RICHIAMO E SOSPENSIONE della FORNITURA dei VACCINI mRNA.
2. REVOCA o SOSPENSIONE dell'AUTORIZZAZIONE ALL'IMMISSIONE IN COMMERCIO.
3. ANNULLAMENTO DI TUTTI I CONTRATTI TRA PFIZER/BIONTECH e CE.
4. RISARCIMENTO delle PERDITE / DANNI: INTERAMENTE A CARICO DEL FABBRICANTE.
5. EMA/CHMP PERSEGUITI PER FALSO

1. REVOCA PEER-REVIEW STATUS
2. REVOCA PREMI NOBEL
3. EMA/CHMP PERSEGUITI PER FALSO

EPAR/RMP di Pfizer/BioNTech afferma: Test di **Genotossicità** & **Cancerogenicità NON** necessari.
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LNP sono da considerarsi ADIUVANTI ?

SI

Il Vaccino **CONTIENE ADIUVANTI**.
EPAR/RMP di Pfizer/BioNTech FALSO
OMISSIONE DOLOSA DI TEST di **Genotossicità** & **Cancerogenicità**

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NO

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FAKE

1.REVOCA PEER-REVIEW STATUS
2. REVOCA PREMI NOBEL
3. EMA/CHMP PERSEGUITI PER FALSO

Approvvigionamento di vaccini anti-COVID-19 nell'UE

Superate le difficoltà iniziali, le dosi necessarie sono state garantite, ma manca un'adeguata valutazione della performance del procedimento d'appalto

10 Un cittadino che abbia subìto effetti indesiderati dovuti a uno dei vaccini contro la COVID-19 acquistati nell'ambito dei contratti può richiedere il risarcimento dei danni al produttore del vaccino. Nel caso in cui la domanda sia accolta, lo Stato membro che ha somministrato il vaccino sarà responsabile del risarcimento della parte lesa e del pagamento delle spese legali sostenute dal produttore (indennizzo) (cfr. *figura 3*). Tale disposizione non si applica se i danni o le perdite sono causati da un comportamento doloso, negligenza grave o mancato rispetto delle buone prassi di fabbricazione dell'UE.

CODICE DI NORIMBERGA

ARTICOLO 5

Non si deve eseguire la sperimentazione se a priori si è a conoscenza che tale sperimentazione possa causare danni o morte…