Sponsor: Novartis Vaccines and Diagnostics S.r.l.

Investigational Product: Trivalent influenza virus vaccine (surface antigen, inactivated, egg-derived) (TIV)

Indication: Prophylaxis against influenza

Protocol Number: V71P5S

Protocol Title: A Phase II, Open Label, Uncontrolled, Multi Center Study to Evaluate Safety and Immunogenicity of AGRIPPAL S1® Surface Antigen, Inactivated, Influenza Vaccine, Formulation 2007-2008, when Administered to Non-Elderly Adult and Elderly Subjects

Phase of Development: Phase II

Study Period:

Date of first enrolment: 18 JUN 2007

Date of last visit: 17 JUL 2007

Methodology:

The trial was designed as a phase II, single-arm, open-label, uncontrolled, multi-center study to evaluate the safety, clinical tolerability and immunogenicity of AGRIPPAL S1 (TIV), formulation 2007-2008 to address the Committee for Medical Products for Human use (CHMP) requirements.

The subjects participating in the study were admitted to the trial on the basis of medical history and a physical examination in order to ensure conformity with the protocol inclusion/exclusion criteria. Vaccine was administered intramuscularly, preferably in the deltoid muscle of the non dominant arm.

Blood samples for immunogenicity assays were collected before vaccination (Day 0) and after 21 (Day 21, window: 20-24) days.

Measurement of antibody titers was performed using the single radial hemolysis (SRH) assay.

In this study, subjects were enrolled into two groups according to age (at least 50 subjects aged 18-60 years should be evaluable; at least 50 subjects aged 61 and over should be evaluable). Subjects were observed for 30 minutes after vaccination on Day 0 for any immediate reactions. Each subject was instructed to complete a diary card for 3 more days post-vaccination to describe local (ecchymosis, induration, erythema swelling and pain) and systemic reactions [fever (i.e., axillary temperature ≥ 38°C), chills/shivering, malaise, myalgia, arthralgia, headache, sweating, fatigue] and was contacted by phone on Day 4 after vaccination to obtain local and systemic reaction data and to determine subject's clinical status. All adverse events (AEs) were collected during Days 0 to 3. All adverse events necessitating a physician's visit or consultation and/or leading to premature study discontinuation, all local and systemic reactions that extend beyond Day 3 and all serious adverse events were collected throughout the trial.

Number of Subjects (planned and analyzed):

125 subjects were enrolled in this study.

Study Centers:

Three sites in Italy

Publication (reference) and/or ClinicalTrials.gov National Clinical Trial (NCT) Number:

NCT00518726

Objectives:

- To evaluate the antibody response to each influenza vaccine antigen, as measured by Single Radial Hemolysis (SRH) at 21 days post-vaccination in non-elderly adult and elderly subjects in compliance with the requirements of the current European Union (EU) recommendations for the evaluation of the immunogenicity for a new formulation of a licensed flu vaccine (CPMP/BWP/214/96).
- To evaluate the safety of the administration of a single intramuscular (IM) injection of AGRIPPAL S1® (TIV) in non-elderly adult and elderly subjects.

Test Product, Dose, Mode of Administration, Lot Number:

The study vaccine, TIV (lot no. 070201), is a subunit influenza vaccine containing highly purified surface antigens obtained from A and B influenza viruses cultivated in embryonate chicken eggs and inactivated with formaldehyde.

A single 0.5mL dose of TIV contained 45µg of viral hemagglutinin, composed of 15µg of each of the strains:

- A(H1N1) strain: IVR-145 (A/Solomon Island/3/2006 - like strain)
- A(H3N2) strain: NYMC X-161B (A/Wisconsin/67/2005-like strain)

- B strain: (B/Malaysia/2506/2004-like strain)

in accordance with the recommendations of the World Health Organization, EU and national Regulatory Agencies for the 2007/2008 season.

The vaccine was packaged in ready to use, single dose syringes. A single 0.5 mL dose of the study vaccine was administered IM in the deltoid muscle of the non-dominant arm.

Duration of Study:

Participation per subjects: 3 weeks

Reference Therapy, Dose, Mode of Administration, Lot Number:

None

Statistical Methods:

Only subjects who contributed evaluable serum samples both before immunization and at 21 days after immunization (acceptable time interval 20-24 days) were to be included in the immunogenicity analysis.

For each age group and each vaccine antigen, geometric mean areas (GMA) were calculated by exponentiating (base 10) the mean of the log (base 10) transformed titers. Day 21 to day 0 geometric mean ratios (GMR) of areas were computed as the geometric mean of the ratios of the day 21 area to the day 0 area of each subject. Percentages of subjects with seroconversion, significant increase or protected were also computed separately for each age group and each vaccine antigen.

Diagnosis and Main Criteria for Inclusion and Exclusion:

Main inclusion criteria:

Individuals eligible for enrollment into this study were male and female adult volunteers who were:

1. 18 years of age or older, mentally competent, willing and able to give written informed consent prior to study entry
2. able to comply with all the study requirements
3. in general good health as determined by:
    - medical history
    - physical examination
    - clinical judgment of the investigator.

Informed consent was obtained for all the subjects before enrollment in the study.

Main exclusion criteria:

Individuals were not to be enrolled into the study if:

1. they had any serious disease (cancer [leukemia, lymphomas, neoplasm], autoimmune disease, advanced arteriosclerotic disease or insulin dependent diabetes mellitus, chronic obstructive pulmonary disease [COPD] that requires oxygen therapy, acute or progressive hepatic disease, acute or progressive renal disease, congestive heart failure);
2. they were hypersensitive to ovalbumin, chicken protein, chicken feathers, influenza viral protein, kanamycin and neomycin sulphate or any other component of the vaccine;
3. they had a history of neurological symptoms or signs, or anaphylactic shock following administration of any vaccine;
4. they had a known or suspected (or have a high risk of developing) impairment/ alteration of immune function;
5. women who were pregnant, or women able to bear children but not willing to practice acceptable contraception for the duration of the trial (21 days); breast feeding women;
6. within the past 12 months, they had received more than one injection of influenza vaccine;
7. within the past 6 months, they had laboratory confirmed influenza disease or received influenza vaccine;
8. within the past 4 weeks they had received another vaccine or any investigational agent;
9. within the past 3 days, they had experienced fever (i.e., axillary temperature $\geq 38°C$);
10. they were taking part in another clinical study;
11. they had any condition which, in the opinion of the investigator, might interfere with the evaluation of the study objective.

Criteria for Evaluation:

Measurement of antibody titers was performed using the single radial hemolysis (SRH) assay.

In order to meet the immunogenicity objective in subjects of age 18-60 years, at least one of the following criteria had to be met ineach of the three virus strains , approximately 3 weeks after vaccination:

- number of seroconversions or significant increase in antibody titer > 40%
- mean geometric increase > 2.5

- percentage of subjects achieving an SRH area ≥25 mm^2 > 70%

In order to meet the immunogenicity objective in subjects aged 61 years and over at least one of the following criteria had to be met in each of the three virus strains, , approximately 3 weeks after vaccination:

- number of seroconversions or significant increase in antibody titer > 30%
- mean geometric increase > 2.0
- percentage of subjects achieving an SRH area ≥25 mm^2 > 60%

Results:

Table 2: Overview of Subject Populations by Age Group

	Number (%) of Subjects	
	TIV	
	(Adults 18-60 Years of Age)	(Elderly >= 61 Years of Age)
Enrolled	63 (100%)	62 (100%)
Immunogenicity – Intention To Treat (ITT)	63 (100%)	62 (100%)
Immunogenicity – Per Protocol (PP)	60 (95%)	60 (97%)
Safety	63 (100%)	62 (100%)

Abbreviation: TIV = Trivalent influenza virus vaccine (surface antigen, inactivated, egg-derived)

Table 3: Summary of Study Terminations - All Enrolled Subjects

	Number (%) of Subjects		
	TIV		
	(Adults 18-60 Years of Age)	(Elderly >= 61 Years of Age)	Total
Total number of subjects enrolled	63	62	125
Completed	60 (95%)	60 (97%)	120 (96%)
Completed protocol	60 (95%)	60 (97%)	120 (96%)
Premature withdrawal	3 (5%)	2 (3%)	5 (4%)
Lost to follow up	1 (2%)	2 (3%)	3 (2%)
Protocol deviation/violation	2 (3%)	0	2 (2%)

Abbreviation: TIV = Trivalent influenza virus vaccine (surface antigen, inactivated, egg-derived)

Table 4: Demographic and Other Baseline Characteristics - All Enrolled Subjects by Age Group

	TIV	
	(Adults 18-60 Years of Age) (N=63)	**(Elderly >= 61 Years of Age) (N=62)**
Age (Years)	39.5±11.7	71.2±5.9
Sex:		
Male	20 (32%)	32 (52%)
Female	43 (68%)	30 (48%)
Ethnic origin:		
Caucasian	62 (98%)	62 (100%)
Hispanic	1 (2%)	-

Categorical parameters: N(%), non-categorical parameters: Mean ± Std

Abbreviation: TIV = Trivalent influenza virus vaccine (surface antigen, inactivated, egg-derived)

Table 5: Immunogenicity Results at Day 0 and Day 21 for Subjects Aged 18-60 Years and Aged 61 Years and Over

		(Adults 18-60 Years of Age) (N=60)							(Elderly >= 61 Years of Age) (N=60)						
Strains		**A/ (H1N1)**		**A/ (H3N2)**		**B**			**A/ (H1N1)**		**A/ (H3N2)**		**B**		
PREVACCINATION (Day 0)															
		n/N	%	n/N	%	n/N	%		n/N	%	n/N	%	n/N	%	
[2]GMT		19		17		33			24		14		39		
[3]95% CI		14-24		12-23		25-43			18-31		10-19		33-47		
[4]Seroprotection Rate		30/60	50	32/60	53	46/60	77		37/60	62	25/60	42	51/60	85	
[3]95% CI		37-63		40-66		64-87			48-74		29-55		73-93		
POSTVACCINATION (Day 21)															
	Requirements	[1]n/N	%	n/N	%	n/N	%	Requirements	n/N	%	n/N	%	n/N	%	
[5]Seroconversion Rate		11/15	73	22/22	100	8/10	80		6/11	55	17/25	68	1/3	33	
[6]Significant increase in antibody titre		19/45	42	19/38	50	15/50	30		10/49	20	15/35	43	13/57	23	
[7]Seroconversion rate or significant increase	> 40%	30/60	**50**	41/60	**68**	23/60	38	>30%	16/60	27	32/60	**53**	14/60	23	
[3]95% CI		37-63		55-80		26-52			16-40		40-66		13-36		
GMT		42		65		59			36		37		48		
[3]95% CI		35-50		59-71		51-67			30-44		29-48		41-57		
Mean GMT Increase	> 2.5	2.22		**3.84**		1.77		>2	1.52		**2.68**		1.23		
[3]95% CI		1.71-2.9		2.76-5.34		1.4-2.25			1.22-1.88		2-3.6		1.11-1.37		
Seroprotection rate	> 70%	52/60	**87**	60/60	**100**	58/60	**97**	>60%	47/60	**78**	49/60	**82**	56/60	**93**	
[3]95% CI		75-94		94-100		88-100			66-88		70-90		84-98		

Bold: CHMP criteria met

Abbreviation: SRH = single radial hemolysis.

[1.] n/N: responders (n) as part of number of subjects of the (sub)population (N), i.e. seroconversion or significant increase.

[2.] GMT: geometric mean titre.

[3.] 95% CI: 95% confidence interval.

[4.] Seroprotection rate: proportion of subjects with a protective titre pre- or postvaccination = SRH-test $\geq 25mm^2$.

[5.] Seroconversion rate: proportion of subjects with antibody increase from prevaccination (seronegative) to postvaccination area $\geq 25mm^2$.

[6.] Significant increase: proportion of subjects with a significant increase in antibody titre, i.e. at least a 50% increase in area.

[7.] Seroconversion rate or significant increase: proportion of subjects with either seroconversion or significant increase in antibody titre.

Table 6: Overview of Solicited Reactions after Administration of TIV, by Age Group.

	Number (%) of Subjects	
	(Adults 18-60 Years of Age) (N=63)	**(Elderly >= 61 Years of Age) (N=62)**
Any	22 (35%)	15 (24%)
Local	15 (24%)	4 (6%)
Systemic	16 (25%)	14 (23%)

Table 7 - Local and Systemic Reactions After the Administration of TIV, by Age Group (0-3 Days Postvaccination).

	Number (%) of Subjects	
	(Adults 18-60 Years of Age) (N=63)	**(Elderly >= 61 Years of Age) (N=62)**
Injection site		
Ecchymosis	1 (2%)	0 (0%)
Erythema	0 (0%)	0 (0%)
Induration	2 (3%)	0 (0%)
Swelling	0 (0%)	0 (0%)
Pain	13 (21%)	4 (6%)
Systemic		
Chills	0 (0%)	3 (5%)
Malaise	5 (8%)	2 (3%)
Myalgia	6 (10%)	4 (6%)
Arthralgia	3 (5%)	7 (11%)
Headache	7 (11%)	3 (5%)
Sweating	6 (10%)	5 (8%)
Fatigue	2 (3%)	5 (8%)
Fever (Temp >=38°C)	0 (0%)	2 (3%)

Table 8: Overview of Unsolicited AEs

None reported.

Table 9: Number (Percentages) of Subjects with Serious Adverse Events by Preferred Term sorted by System Organ Class

None reported.

Table 10: Number (Percentages) of Subjects with Unsolicited Adverse Events Reported by ≥ 5% of Subjects by Preferred Term sorted by System Organ Class

None reported.

Conclusion:

Based on the reported results, the 2007/2008 TIV S1 influenza vaccine had a good immunogenicity and safety profile and met the CHMP criteria for approval of influenza vaccines in Europe.

Date of Clinical Trial Report: 25 JUL 07