

Molecular Study Using Real Time PCR to Detect Influenza Viruses (Flu A, Flu B and RSV) in Patients at Ramadi Hospital

Abbas O. Aljanabi¹, Mohammed Amer Fayyadh², Aseel Khalid Hameed³, Najeeb Mohammed Hussein⁴

¹Assistant Prof Microbiology Department, College of Medicine, University Of Anbar, ²Lecturer, Collage of Medicine , University of Fallujah,Iraq, ³Lecturer, Al Ramadi Health Center, Ministry of Health, Iraq,

⁴Assistant Prof. Department of Food Sciences , College of Agriculture , University Of Anbar, Iraq

Abstract

Background: Because of the increasing incidence of infections with different types of influenza of various types, type A, B, and SRV, this study included investigating the type of infection in influenza in women and men in different age groups and making a comparison between them. **Methodology:** 90 blood samples were collected, including 30 samples for men 30 samples for women showing clinical symptoms of infection, and 30 samples from people with clinical symptoms under control. These samples were placed in tubes containing an anticoagulant to ensure that blood does not clot DNA/RNA extraction kit, CerTest, Spanish Company and detection by Flu A, Flu B and RSV, CerTest, Spanish Company by using real time PCR. **Result:** Table 1 shows the investigation of some types of influenza in women who showed symptoms of infection and who are between 20 to 40 years old, where the frequency of infection in women between the ages of 20-30 years was high with types Flu A, SRV compared with control and did not show infection with type B for the same Age group . As for women aged between 30-40 years, the frequency of type B infection was high compared to control, and there were no infections with Flu a and SRV for the same age group .

Keyword: Real time PCR , Flu A, Flu B and RSV

Introduction

There are three sorts of seasonal influenza A, B, and C. Type A influenza viruses additionally department into subtypes in accordance to combos of two unique proteins, haemagglutinin (H) and neuraminidase (N), placed on the floor of the virus. (1,2) Among the many influenza A virus subtypes,

the two subtypes A (H1N1) and A (H3N2) are currently circulating in humans. (3,4) The communicable influenza virus A (H1N1) is referred to as A (H1N1) pdm09 as it brought about pandemic influenza in In 2009 it due to this fact changed the seasonal influenza A (H1N1) virus that used to be circulating earlier than 2009. Influenza viruses by myself are recognized to purpose pandemics (5,6) Influenza B viruses can be divided into two most important training (strains) referred to as the B / Yamagata stress and the B / Victoria strain. Type B influenza viruses are no longer categorized as

Corresponding:

Najeeb Mohammed

dr.najeb@uoanbar.edu.iq

subtypes^(7,8) Influenza viruses of kind A and B flow into and motive outbreaks and epidemics. ^(9,10) For this reason, the applicable lines of influenza viruses A and B are covered in seasonal influenza vaccines. Influenza C virus is detected solely in uncommon instances and typically motives slight infections and for this reason has much less extreme public fitness effects.^(11,12) Seasonal flu is characterised by way of a unexpected excessive physique temperature, cough (usually dry), headache, muscle and joint pain, extreme nausea (malaise), sore throat, and runny nose. You can have a extreme cough that lasts two weeks or more. Most sufferers unravel their fever and different signs and symptoms inside one week besides desiring scientific attention. However, influenza can reason extreme sickness or even dying if it impacts one of the high-risk businesses (see below).^(13,14) The duration between the acquisition of the contamination and the onset of the disease, recognized as the incubation period, lasts about two days. ⁽¹⁵⁾

Methodology

Samples collection

90 blood samples were collected, including 30 samples for men 30 samples for women showing clinical symptoms of infection, and 30 samples from people with clinical symptoms under control.

These samples were placed in tubes containing an anticoagulant to ensure that blood does not clot

RNA Extraction and Detection

DNA/RNA extraction kit, CerTest, Spanish Company and detection by Flu A, Flu B and RSV, CerTest, Spanish Company by using real time PCR ..⁽¹⁶⁾

Reagents and equipment to be supplied by the user The ⁽¹⁷⁾

Creating PCR test programmer for VIASURE Flu A, Flu B & RSV Real Time PCR Detection kit . ⁽³⁾

Result and Discussion

All flu subtypes contain different strains of the influenza virus. Not all strains infect people. Subtypes of influenza A viruses that currently appear in people are H1N1 and H3N2 strains. Each year the influenza vaccine contains a variety of these two strains and influenza B. In most cases, the body’s immune system will fight the virus itself. But some people will experience additional complications. These complications are more common in older adults and those with conditions that affect their immune systems. Taking immunosuppressant medications may also increase the risk of complications.

Table (1) Screening for the three types of influenza (Flu A, Flu B, SRV) in women whose ages range from 20 to 40 years old

Descriptive Statistics				
Dependent Variable: Number of Virus copy				
age of woman patient	type of Flu	Mean	Std. Deviation	N
20-30 year	FLU A	34.60	8.081	5
	FLU B	3.40	1.140	5
	RSV	35.00	5.874	5
	Total	24.33	16.238	15

Cont... Table (1) Screening for the three types of influenza (Flu A, Flu B, SRV) in women whose ages range from 20 to 40 years old

30-40 year	FLU A	3.80	.837	5
	FLU B	39.60	16.682	5
	RSV	3.00	.707	5
	Total	15.47	19.799	15
control	FLU A	2.80	.837	5
	FLU B	3.20	.837	5
	RSV	3.40	1.140	5
	Total	3.13	.915	15
Total	FLU A	13.73	15.890	15
	FLU B	15.40	19.845	15
	RSV	13.80	15.848	15
	Total	14.31	16.917	45

Table 1 shows the investigation of some types of influenza in women who showed symptoms of infection and who are between 20 to 40 years old, where the frequency of infection in women between the ages of 20-30 years was high with types Flu

A, SRV compared with control and did not show infection with type B for the same Age group . As for women aged between 30-40 years, the frequency of type B infection was high compared to control, and there were no infections with Flu a and SRV for the same age group.

Table (2) ANOVA table for Screening of the three types of influenza (Flu A, Flu B, SRV) in women whose ages range from 20 to 40 years old

Tests of Between-Subjects Effects					
Dependent Variable: Number of Virus copy					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	11058.444a	8	1382.306	32.457	.000
Intercept	9216.356	1	9216.356	216.403	.000
age	3400.844	2	1700.422	39.926	.000
Flu type	26.711	2	13.356	.314	.733
age * Flu type	7630.889	4	1907.722	44.794	.000
Error	1533.200	36	42.589		
Total	21808.000	45			
Corrected Total	12591.644	44			

a. R Squared = .878 (Adjusted R Squared = .851)

Table 2: Analysis of variance for the detection of the three types in women whose age ranges between 4-20 years, as it was noted that there are significant differences between influenza type A for women within the age group 20-30 years compared to control and compared to the age group between

30-40 years while the second type From influenza B, there were clear significant differences in the large age groups, located between 30-40, compared to control and also compared to the younger age groups.

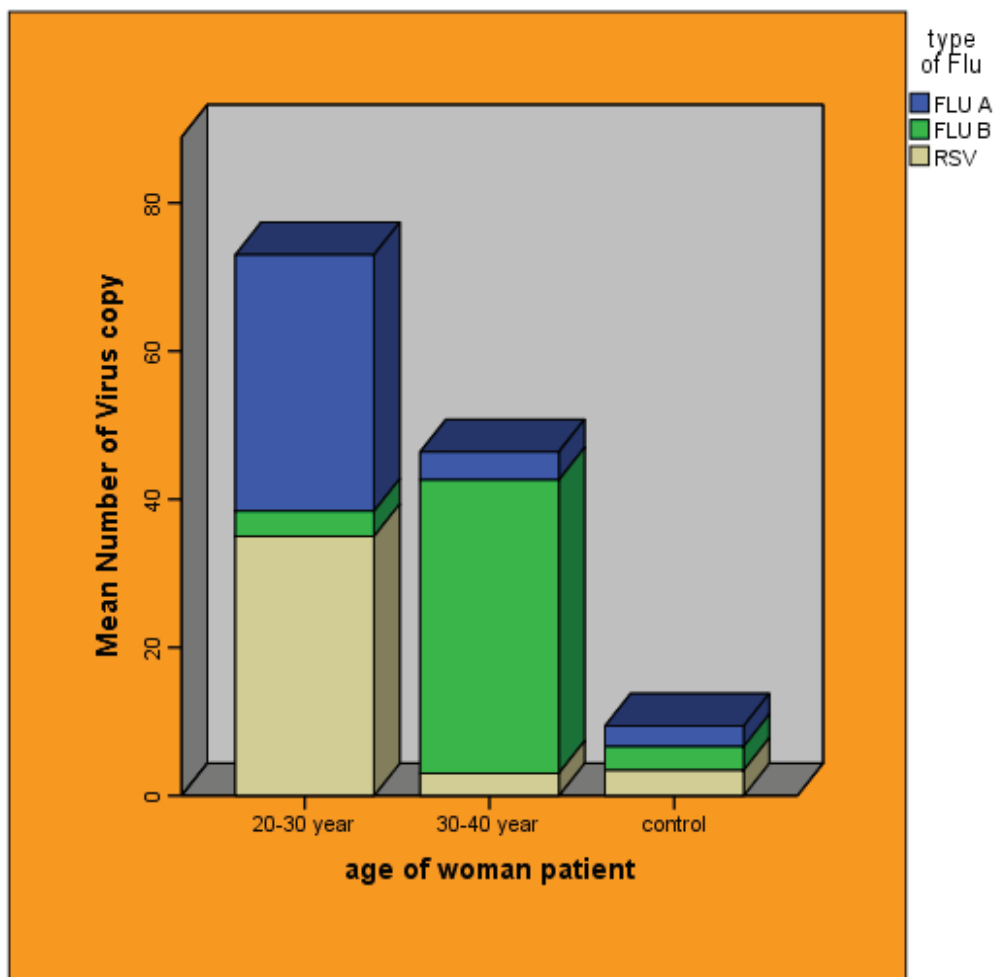


Figure 1 detection of three types of influenza (Flu A, Flu B, SRV) in women whose ages range from 20 to 40 years old

Table (3) Screening for the three types of influenza (Flu A, Flu B, SRV) in men whose ages range from 20 to 40 years old

Descriptive Statistics				
Dependent Variable: Number of Virus copy				
age of men patients	type of Flu	Mean	Std. Deviation	N
20-30 year	FLU A	27.00	5.958	5
	FLU B	3.80	.837	5
	RSV	4.40	1.517	5
	Total	11.73	11.659	15
30-40 year	FLU A	33.80	7.791	5
	FLU B	3.60	1.140	5
	RSV	4.00	1.581	5
	Total	13.80	15.256	15
control	FLU A	4.60	1.517	5
	FLU B	6.40	1.140	5
	RSV	2.80	.837	5
	Total	4.60	1.882	15
Total	FLU A	21.80	13.960	15
	FLU B	4.60	1.639	15
	RSV	3.73	1.438	15
	Total	10.04	11.589	45

Epidemiological data from Hong Kong showed that adult men are more at risk of hospitalization due to influenza. An American study revealed that men died more often from influenza compared to women of the same age, regardless of other underlying diseases such as heart, cancer, and organs. It was found that this gap in immunity, may be caused by hormonal changes, as the male hormone testosterone suppresses the immune system, while the hormone estradiol protects it. It is not usually

known that testosterone is an immunosuppressive, but one study found that men who have levels The higher testosterone they have had less response than the vaccination antibody. Table 3 shows the investigation of the three types of influenza in men whose age ranges between 20-40 years. It was found that there are infections in men whose ages range from 20-30, as well as men between the ages of 30-40 years in influenza type A and no infections have appeared. The other is type B and SRV.

Table (4) ANOVA table of Screening for the three types of influenza (Flu A, Flu B, SRV) in men whose ages range from 20 to 40 years old

Tests of Between-Subjects Effects					
Dependent Variable: Number of Virus copy					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5480.711a	8	685.089	57.463	.000
Intercept	4540.089	1	4540.089	380.809	.000
age	698.978	2	349.489	29.314	.000
Flu type	3114.978	2	1557.489	130.637	.000
age * Flu type	1666.756	4	416.689	34.951	.000
Error	429.200	36	11.922		
Total	10450.000	45			
Corrected Total	5909.911	44			

a. R Squared = .927 (Adjusted R Squared = .911)

Table 4 Analysis of variance shows that there are significant differences in the incidence of influenza A type in the age group 20-30 and 30-40 years compared to other types of influenza and compared to control and there are no significant differences for type B and type SRV. Available research indicates that men suffer worse from colds and influenza than women, calling for “better

quality research” to prove this. Hospitalization rates are consistently higher for younger (before puberty) and older males (over 65 years of age). During a woman’s reproductive years, she often suffers from more severe illnesses, partly because the flu is worse for pregnant women, but also because women develop a greater inflammatory response to the flu.

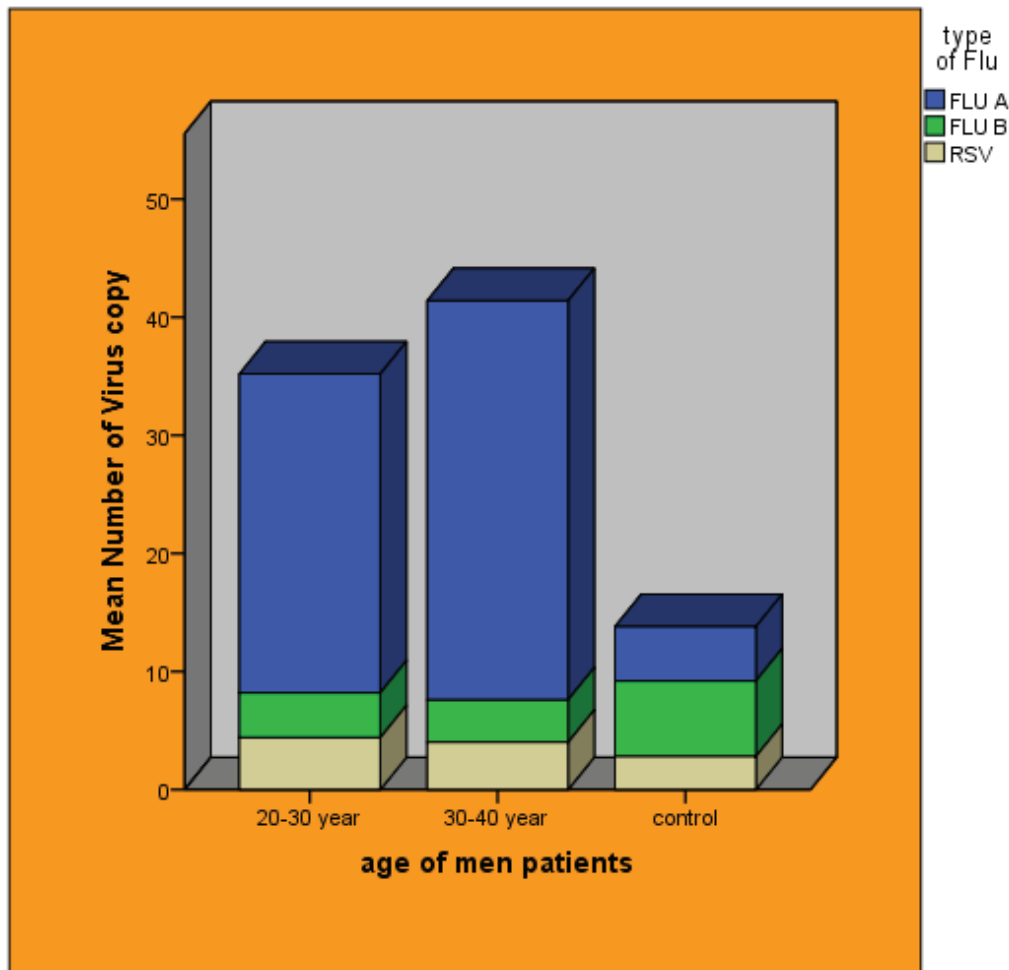


Figure 2 Screening for the three types of influenza (Flu A, Flu B, SRV) in men whose ages range from 20 to 40 years old

Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both MOH and MOHSER in Iraq

Conflict of Interest: None

Funding: Self-funding

References

- Nidhi P, Rolta R, Kumar V, Dev K, Sourirajan A. Synergistic potential of Citrus aurantium L . essential oil with antibiotics against Candida albicans. J Ethnopharmacol [Internet]. 2020;262(April):113135. Available from: <https://doi.org/10.1016/j.jep.2020.113135>
- Publication A. Simultaneous Volumetric and Functional Assessment of the Right Ventricle in Hypoplastic Left Heart Syndrome After Fontan Palliation, Utilizing 3-Dimensional Speckle-Tracking Echocardiography. 2020;
- Bellini F, Michels L, Cristina A, Amaral DC, Carvalho-filho RJ De, Vieira GDA, et al. Hepatitis C treatment of renal transplant and chronic kidney disease patients : efficacy and safety of direct-acting antiviral regimens containing sofosbuvir. 2020;45–9.
- Muthukutty B, Arumugam B, Chen S, Ramaraj SK. Pr pr oo f. J Hazard Mater [Internet]. 2020;124745. Available from: <https://doi.org/10.1016/j.jhazmat.2020.124745>
- Cai Y, Zhang J, Xiao T, Peng H, Sterling SM. Distinct conformational states of SARS-CoV-2 spike protein. 2020.
- Chen G, Zhao J, Ning Q, Chen G, Wu D, Guo W, et al. Clinical and immunological features of severe and moderate coronavirus disease 2019 Clinical and immunological features of severe and moderate coronavirus disease 2019. 2020;130(5):2620–9.
- Msph CLO, Binnicker MJ, Poland GA. Potential impact of contaminated bronchoscopes on novel coronavirus disease (COVID-19) patients. 2020;862–4.
- Xie C, Jiang L, Huang G, Pu H, Gong B, Lin H, et al. International Journal of Infectious Diseases Comparison of different samples for 2019 novel coronavirus detection by nucleic acid amplification tests. Int J Infect Dis [Internet]. 2020;93:264–7. Available from: <https://doi.org/10.1016/j.ijid.2020.02.050>
- Microbes E. Diagnosis and clinical management of severe acute respiratory syndrome Coronavirus 2 (SARS- CoV-2) infection : an operational recommendation of Peking Union Medical College Hospital (V2 . 0). 2020;1751.
- No Title. 2020;(169).
- An P, Zhun Y, Yang L. Biochemical indicators of coronavirus disease 2019 exacerbation and the clinical implications. Pharmacol Res [Internet]. 2020;159(1):104946. Available from: <https://doi.org/10.1016/j.phrs.2020.104946>
- Batlle D, Wysocki J, Satchell K. Soluble angiotensin-converting enzyme 2 : a potential approach for coronavirus infection therapy ? 2020;0:543–5.
- Li Y, Deng W, Xiong H, Li H, Chen Z, Nie Y, et al. Immune - related factors associated with pneumonia in 127 children with coronavirus disease 2019 in Wuhan. 2020;(June):2354–60.
- Chen X, Zheng F, Qing Y, Ding S, Yang D, Lei C, et al. Title : Epidemiological and clinical features of 291 cases with coronavirus disease 2019 in areas adjacent to Hubei , China : a double-center observational study. 2020;
- Hussein, N. M., Atea, A. M., Humide, A. O., Abdullah, Q. K., & Hardan, S. M.. ISOLATION AND DIAGNOSIS OF BACTERIA CAUSING URINARY TRACT INFECTION IN CHILDREN. Systematic Reviews in Pharmacy.2020 11(1), 76-79]
- Musa, F. H., Khaleel, R. W., & Hussein, N. M. Effect of some plant extracts on the Pyocyanin Production from Pseudomonas Aeruginosa

which Isolated from clinical samples. In IOP Conference Series: Materials Science and Engineering .2020.(Vol. 870, No. 1, p. 012041). IOP Publishing

17. Abdullah, Q. K., Sharad, A. A., Hamdi, R. F., Khammas, R. K., & Mohammed Hussein, N.

Effect of celiac disease on humoral immune response and some of the blood variables in children. In AIP Conference Proceedings .2020. (Vol. 2213, No. 1, p. 020240). AIP Publishing LLC