



A Study of Changes in Lymphocyte-to-Monocyte Ratio in Subjects with Covid-19 Vaccine-Induced Toxicity in Comparison to Those Found in Symptomatic Covid-19 Infected Patients

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Abstract

The acute and severe decline in lymphocyte-to-monocyte ratio (LMR), due to both lymphocyte decline and monocyte increase, represents one of the most predictive biomarkers of the severity of Covid-19 infection, while its profile in Covid-19 vaccine toxicity has not been adequately investigated. The present study was performed to evaluate changes in LMR under Covid-19 vaccination, by comparing the results to those observed in a group of hospitalised patients for Covid-19 infection. The study included 50 hospitalised patients, 16 of whom required mechanical ventilation, and 5 of them died from respiratory failure. Abnormally low values of LMR were present in 32/50 (64%) at the time of the hospitalisation, which progressively returned within the normal range at the time of demission. In contrast, a persistent progressive decline in LMR was observed in died patients. Moreover, changes in LMR were evaluated in 14 consecutive subjects with important Covid-19 vaccine-induced toxicity, by comparing the results to those found in 30 subjects, who had no side-effect after Covid-19 vaccination. LMR mean values significantly decreased after vaccination in the only subjects with vaccine-related toxicity, whereas no significant change was observed in those, who had no vaccine-induced side-effect. The results of this preliminary study would suggest that the alterations in LMR values occurring under toxicity due to Covid-19 vaccination are like to those observed in symptomatic Covid-19 infected patients, by consisting of an important decline in LMR values.

Keywords: Covid-19 infection; Covid-19 vaccine; Lymphocyte-to-monocyte ratio; Spike protein

Introduction

According to the experimental and clinical results reported in the literature, the severe physiopathology of Covid-19 infection would be the results of three fundamental pathogenetic events, consisting of: 1) the down-regulation of ACE2 receptor expression in response to the link of the spike protein to ACE2 itself as its receptor, with a following diminished production of angiotensin 1-7 (Ang 1-7) and a concomitant enhanced production of angiotensin II (Ang II), which stimulates IL-17 secretion (2); 2) an enhanced IL-17 secretion which furtherly

stimulates Ang II production and promotes the secretion of macrophage-related inflammatory cytokines, including IL-6 and TNF-alpha whereas ACE2 expression and the consequent Ang 1-7 secretion are inhibited by IL-17 (2); 3) an acute endocannabinoid deficiency due to an activation of the fatty acid amide hydrolase (FAAH), the enzyme involved in cannabinoid degradation and the consequent further increase in IL-17 levels, since IL-17 secretion is under an inhibitory cannabinoid content [1-8]. The major severe symptoms of Covid-19 infection, including acute respiratory distress syndrome (ARDS), and systemic thrombosis, would be due the enhanced secretion of both

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Ang II and IL-17, since either Ang II, or IL-17 have appeared to induce direct lung damage, cardiotoxicity, neurotoxicity, and thrombotic events as well as to a concomitant Ang 1-7 deficiency which has been proven to exert anti-thrombotic effects, a cardio-neuro-protective activity, and a prevention of lung inflammatory damage [9-12]. Recently, however, Ang 1-7 blood levels have appeared to be increased in patients with severe Covid-19 infection [13]. Therefore, changes in Ang II and Ang 1-7 secretion in Covid-19 infection need to be better investigated and defined, particularly in relation to IL-17 secretion, which would represent the main cytokine responsible for Covid-19-related exaggerated inflammatory response. In any case, the pathogenesis of Covid 19 disease could be synthetically interpreted as an acute and severe changes in renin-ACE-ACE2-Ang II Ang 1-7 functional axis, who's functionless is fundamental for the human status of health [14]. The rare Covid 19 vaccine-related toxicity also could be due to similar mechanisms induced by spike protein itself. As far as Covid 19 vaccine-induced toxicity is concerned, at present the main side-effects have appeared to consist of fever, asthenia, myalgia, headache, chills, nausea and vomiting, diarrhoea, and palpitation, with a trouble temporally to perform the activities of daily living in about 12% subjects for some days with only few differences among the different types of vaccine [15,16]. From an immune pathological point of view, despite the complexity of immune and cytokine mechanisms involved in Covid 19 infection, the end-result has been proven to synthetically consist of an acute decline in lymphocyte count in association with a concomitant increase in neutrophil and monocyte numbers [17]. Moreover, the increase in neutrophil-to-lymphocyte ratio (NLR) has appeared to be associated with a more severe clinical course. In addition, the decline in lymphocyte-to-monocyte ratio (LMR) has been proven to be more appropriate in predicting the severity of Covid 19 infection [18]. Then, LMR could represent the main biomarker to clinically monitor the evolution of Covid 19 disease, since it would synthetize within itself the great number of immune cell and cytokine alterations occurring during Covid 19 infection, as well as in other systemic diseases [19]. On these bases, a preliminary study was planned to evaluate LMR behaviour in response to Covid 19 vaccination and its eventual toxicity, by comparing the results with those observed in a group of Covid 19 infected patients with important symptoms, who had been hospitalized and investigated before the introduction of Covid 19 people vaccination.

Patients and Methods

The phase 2 study included 14 consecutive subjects (M/F: 10/4; median age: 62 years; range 22-91), who had important side-effects after the first Covid-19 vaccine subcutaneous injection (Pfizer: 9; Astrazeneca: 3; Moderna: 2). the type of vaccine was

established according to the dispositions of Italian Health Ministry in relation to the age of subjects. The results were compared to those found in a control group of 30 subjects (M/F: 18/12; median age 56 years, range 35-84), who had no toxicity after Covid 19 vaccination (Pfizer: 18; Astrazeneca: 7; Moderna: 5). the study was performed during the first of the three planned doses of vaccination. The subjects, who had a previous Covid 10 infection, were not included in the study. Covid- 19 vaccine-induced side-effects were considered as an important toxicity in the presence of one or more of the following symptoms: fever greater than 39° C for at least two days, respiratory difficulty, thrombotic events, fatigue, or myalgia for more than 7 days, severe diarrhoea, autoimmune or allergic reactions, and cardiac arrhythmia. The results were also compared to those observed in 50 patients, who were hospitalized for Covid 19 infection before the introduction of vaccination (M/F: 32/18; median age 71 years, range 33-90). Among the hospitalized patients for Covid 19 infection, 16/50 (32%) required mechanic ventilation because of important respiratory failure, and 5/16 (31%) died from ARDS, systemic thrombosis, and multi-organ failure. To evaluate changes in LMR, in the group of subjects who received Covid 19 vaccine, venous blood samples were collected before vaccination, and at 5 and 15 days after the first vaccine injection. In patients hospitalised for Covid 19 infection with important systemic symptoms, venous blood samples were drawn at hospitalisation moment, and at daily intervals depending on the severity of disease, by considering the maximal LMR variation on study. Normal values obtained in our laboratory (95% confidence limits) were, as follows: lymphocytes greater than 1,500/mm³, monocytes less than 450/mm³, and LMR greater than 2.1. Data were reported as mean +/-SE, and statistically analysed by the chi-square test, Student's t test, and analysis of variance, as appropriate.

Results

The main side-effects observed in the 14 subjects with vaccine-related toxicity consisted of fever greater than 39° C for more than three days with asthenia and myalgia in 6/14 (43%), thrombosis in 2/14 (14%), and important respiratory failure requiring mechanic ventilation in the remaining 6 patients (43%). However, no subject died from Covid 19 vaccine-induced toxicity. In subjects who had no important toxicity after vaccination, no significant changes in lymphocyte and monocyte mean numbers occurred. Then, LMR mean values did not change after vaccination. On the contrary, in subjects with vaccine-induced side-effects, lymphocyte and monocyte mean numbers significantly decreased and increased after vaccine, respectively, and they returned to the pre-vaccination values within 15 days after vaccination. Then, LMR mean values significantly decreased after vaccination with respect to the pre-vaccination

values ($P < 0.05$). Lymphocyte, monocyte, and LMR mean values observed in subjects with vaccine-induced side-effects are illustrated in (Figure 1). Moreover, by considering the immune variations in relation to the severity of vaccine-induced toxicity, as illustrated in patients who did not require the mechanic ventilation, monocyte mean count significantly enhanced after vaccination, while lymphocyte mean number decreased but not in a significant manner, and also LMR decline was not statistically significant. On the contrary, in intubated patients, while monocyte increase was not significant, lymphocyte mean number significantly decreased after vaccination, and on the same way, LMR mean values significantly diminished after vaccination (Figure 2). In any case, lymphocyte, monocyte, and LMR mean values became within the normal range within 15 days after vaccination.

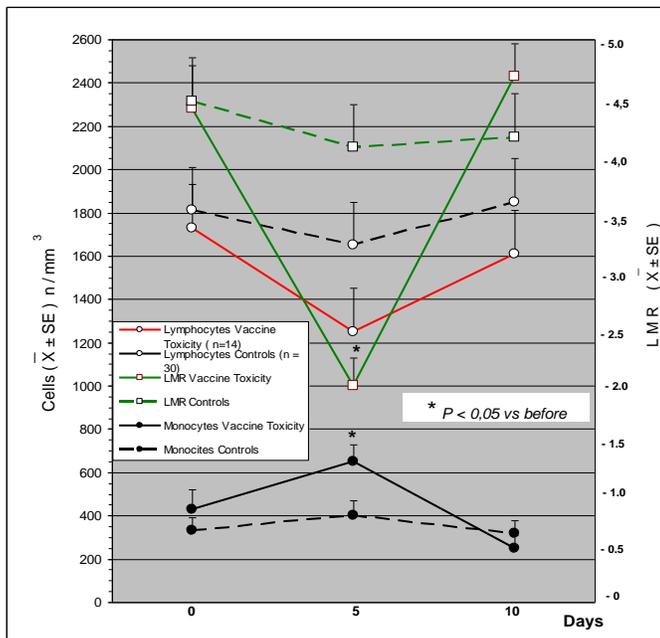


Figure 1: Changes in lymphocyte, monocyte, and lymphocyte-to-monocyte ratio (LMR) mean values in 14 subjects with Covid 19 vaccine-induced side-effects and in controls ($n=30$).

On the other hand, in the group of patients hospitalized for Covid 19 infection before the introduction of vaccination, lymphocytopenia and monocytosis occurred in 39/50 (78%), and in 23/50 (46%) patients, respectively at the time of the hospitalisation. Therefore, an abnormally low LMR was present in 32/50 (64%) patients. Moreover, in the 34 patients who did not require mechanic ventilation, lymphocyte and monocyte counts progressively increased and decreased, respectively, and the mean values of lymphocytes and monocytes observed at the time of demission were respectively significantly higher ($P < 0.01$) and lower ($P < 0.05$) than those found at the time of the hospitalisation. In addition, in both patients, who did not require mechanic

ventilation, or who were recovered after mechanic ventilation ($n=11$), LMR values observed at the demission were within the normal range, whereas in patients who died under mechanic ventilation LMR mean values progressively declined in a statistically significant manner with respect to those seen at the time of the hospitalisation ($P < 0.01$ vs before and the other group). Shows changes in mean values of lymphocytes, monocytes and LMR in Covid 19 infected patients in relation to their clinical evolution (Figure 3).

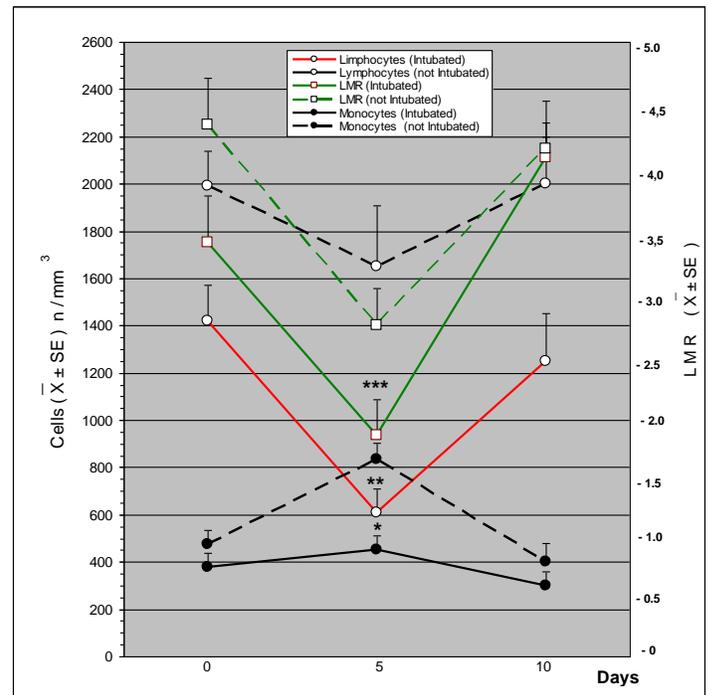


Figure 2: Changes in lymphocyte, monocyte, and lymphocyte-to-monocyte ratio (LMR) in subjects with Covid 19-induced toxicity, who required ($n=6$) or not required ($n=8$) mechanic ventilation. (* $P < 0,01$ vs before, $P < 0,001$ vs non-intubated; ** $P < 0,01$ vs before, $P < 0,05$ vs Intubated; *** $P < 0,01$ vs before, $P < 0,05$ vs non-Intubated).

Discussion

The results of this preliminary study seem to suggest that the mechanisms involved in Covid 19 vaccine-induced side-effects are similar to those occurring in Covid 19 disease, by consisting of a decline in lymphocyte count in association with a concomitant increase in monocyte number, with, however, some differences about both lymphocyte and monocytes behaviours, since lymphocytopenia is more typical of Covid-infection and vaccine-induced severe toxicity, while monocyte increase seems to occur more frequently in subjects with Covid-19 vaccine-induced mild side-effects. In any case, from a clinical point of view, both Covid 19 disease and Covid 19 vaccine-induced

toxicity may be monitored by evaluating LMR values, since an evident decline in LMR values is connected to either the severity of Covid 19 disease, or to severe side-effects following Covid 19 vaccination. This finding is not surprising since the complications of both Covid 19 disease and vaccination would be mainly due to the direct action of spike protein itself [1-4].

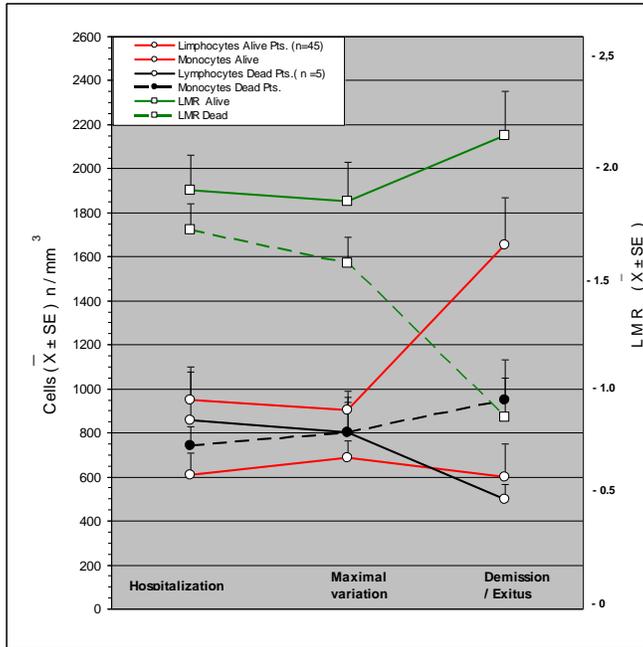


Figure 3: Changes in mean values of lymphocyte count, monocyte number, and lymphocyte-to-monocyte ratio (LMR) in 50 patients hospitalised for Covid-19 infection, who required no mechanic ventilation ($n=34$), who were recovered after mechanic ventilation ($n=11$), or who died under mechanic ventilation ($n=5$).

Further studies, however, by detecting Ang 1-7 blood levels with respect to those of Ang II, IL-17 and FAAH blood concentrations in both Covid 19 infected patients and healthy subjects under vaccination, will be required to establish whether both Covid 19 disease complications and Covid 19 vaccine-induced side-effects may be due to an Ang 1-7 and cannabinoid deficiencies in association with a concomitant increase in IL-17 and Ang II endogenous production. Moreover, further studies will be required to identify possible differences in the immune mechanisms and cytokine response related to the different Covid 19 vaccines. Finally, further clinical investigations will be needed to evaluate which relation exists between changes in LMR ratio after vaccination and anti-Covid 19 antibody production.

References

- Verdecchia P, Cavallini C, Spanevello A, Angeli F. The pivotal link between ACE2 deficiency and SARS-CoV-2 infection. *Eur J Intern Med.* 2020; 76: 14-20.
- Madhur MS, Lob HE, McCann LA, Iwakura Y, Blinder Y, Guzik TJ, et al. Interleukin-17 promotes angiotensin II-induced hypertension and vascular dysfunction. *Hypertension.* 2010; 55: 500-507.
- Mahallawi WH, Khabour OF, Zhang O, Makhdoum HM, Suliman BA. MERS-CoV infection in humans is associated with a pro-inflammatory Th1 and Th17 cytokine profile. *Cytokine.* 2018; 104: 8-13.
- Casillo GM, Mansopur AA, Raucci F, Saviano A, Mascolo N, Iqbal AJ, et al. Could IL-17 represent a new therapeutic target for the treatment and/or management of COVID-19-related respiratory syndrome? *Pharmacol Res.* 2020.
- Kryczek I, Wei S, Vatan L, Escara-Wilke J, Szeliga W, Keller ET. Cutting edge: opposite effects of IL-1 and IL-2 on the regulation of IL-17+T cell pool IL-1 subverts IL-2 mediated suppression. *J Immunol.* 2007; 179: 1423-1426.
- Lissoni P, Rovelli F, Monzon A, Messina G, Porta E, Porro G, et al. COVID-19 disease as an acute angiotensin 1-7 deficiency: a preliminary phase 2 study with angiotensin 1-7 in association with melatonin and cannabidiol in symptomatic COVID 19-infected patients. *J Infectology.* 2020; 3: 13-16.
- Grotenhermen F. Pharmacology of cannabinoids. *Neuroendocrinol Lett.* 2004; 25: 14-23.
- Nagarkatti P, Pandey R, Rieder SA, Hegde VL, Nagarkatti M. Cannabinoids as novel anti-inflammatory drugs. *Future Med Chem.* 2009; 1: 1333-1349.
- Robert M, Miossec P. Effects of interleukin-17 on the cardiovascular system. *Autoimmune Rev.* 2017; 16: 984-991.
- Cheng H, Wang Y, Wang GQ. Organ-protective effect of angiotensin-converting enzyme 2 and its effect on the prognosis of COVID-19. *J Med Virol.* 2020; 92: 726-730.
- Osman IO, Melenotte C, Brouqui P, Million M, Lagier JC, Parola F. Expression of ACE2, soluble ACE2, Angiotensin I, Angiotensin II and Angiotensin-(1-7) is modulated in COVID-19 patients. *Front Immunol.* 2021.
- Capettini LS, Montecucco F, Mach F, Stergiopoulos N, Santos RA, da Silva RF. Role of renin-angiotensin system in inflammation, immunity, and aging. *Curr Pharm Des.* 2012; 18: 963-970.
- Reindl-Schwaighofer R, Hodlmoser S, Eskandary F, Poglitsch M, Bondeman D, Strassl R. ACE2 elevation in severe COVID-19. *Am J Respir Crit Care Med.* 2021; 203: 1191-1196.
- Imanpour H, Rezaee H, Nouri-Vaskeh M. Angiotensin 1-7: a novel strategy in COVID-19 treatment. *Adv Pharm Bull.* 2020; 10: 488-489.
- Kadali RAK, Janagama R, Peruru S, Malayala SV. Side effects of BNT162b2 mRNA COVID-19 vaccine: A randomized, cross-sectional study with detailed self-reported symptoms from healthcare workers. *Int J Infect Dis.* 2021; 106: 376-382.
- Meo SA, Bukhari IA, Akram J, Meo AS, Klonoff DC. COVID-19 vaccines: comparison of biological, pharmacological characteristics and adverse effects of Pfizer/BioNTech and Moderna Vaccines. *Eur Rev Med Pharmacol Sci.* 2021; 25: 1663-1669.



SUNTEXT REVIEWS

17. SeyitM, Auci E, Nar R, SenolH, Yilmaz A,OzenM, Oskay A, Aybek H. Neutrophil to lymphocyte ratio, lymphocyte to monocyte ratio and platelet to lymphocyte ratio to predict the severity of COVID-19. *Am J EmergMed.* 2021; 40: 110-114.
18. Lissoni P, Rovelli F, Monzon A, Privitera C, Messina G, Porro G. Evidenceofabnormallylowlymphocyte-to-monocyte ratio in Covid-19-induced severe acute respiratory syndrome. *J Immuno Allerg.* 2020; 1: 1-6.
- Lissoni P, Brivio F, Fumagalli L, Rovelli F, Colciago M, Messina G. Howto modulate the immunoinflammatory biological response in cancer, autoimmunity and viral infections by a psycho-neuroimmune approach. *Nov Appro in Can Study.* 2021; 5: 540-549.