Polymorphisms of IFN-γ T/A +874 Gene and Relationship with COVID 19 in Iraqi Population

Anwar Abed Nasser Dhabaan*1 and Mohammad Hussein Alwan²
¹²Al-Iraqia University / Collage of Education, Iraq
dr.anwar.a.nasser@gmail.com

ABSTRACT
This study included 80 blood specimens. Fifty samples collected from COVID 19 with age ranged between 20-75 years, and 30 blood specimens collected from healthy as a control sample with age ranged between 91-63 years. The polymorphism of IFN-γ T/A +874 gene, which amplified by using amplification refractory mutation system (ARMS-PCR) was showed high percentage of A allele frequency in COVID 19 patients sample in comparison with T allele frequency, and the A allele revealed as etiological fraction with risk by having COVID 19 disease, whereas the T allele showed high frequency from the A allele frequency in control specimen, and the A allele revealed as preventive fraction from infection by this disease. The AA and TA genotypes revealed as etiological fraction with risk by having COVID disease, whereas the TT genotype revealed as preventive fraction with risk by having COVID 19 disease. Our findings demonstrate that the IFN-γ Gene T/A +874 gene polymorphism may represent a significant risk factor for COVID 19 in Iraqi population and there is association between the IFN-γ Gene T/A +874 polymorphism and COVID 19 patients.

Keywords: Interferon-gamma gene, polymorphism and COVID 19

1. Introduction
Coronaviruses are a large and widespread family and infect a variety of organisms, including mammals and birds. The first strain was discovered in 1931 that causes infectious bronchitis to birds (IBV) and is considered the first corona virus to be discovered [1]. The Covid 19 virus is a coronavirus and appears under an electron microscope in a rough spherical or multifaceted crystalline shape, and has elevations in the membrane in the shape of a crown or wreaths [2]. The virus has the viral genome, which is RNA, and has a molecular weight of approximately 26,000 to 32,000 base pairs, and these viruses are among the broadest viruses that carry RNA [3]. The severe acute respiratory syndrome (SARS-CoV-2) virus appeared for the first time in Wuhan, China, in December of 2019, and it is a type of highly pathogenic virus (HCoV) that infects living organisms, including humans, and poses a great threat to public health [4]. Symptoms or signs of infection with the COVID-19 virus varied from person to person, but all symptoms were receding by high temperature (fever), cough, shortness of breath, fatigue, muscle pain, headache, nausea or vomiting, sore throat, congestion or runny nose, diarrhea, loss of smell and loss Taste in addition to neurological and cardiac complications [5]. The methods of diagnosing the virus are different, but the most important way to diagnose the virus is the (PCR) method and its main concern is RT-PCR, reverse transcription and real-time reverse transcription [6]. Cell motility plays an important role in responding to viral infections, and the rapid and coordinated innate immune response is the first line of defense against this infection, and unregulated and excessive immune
responses may cause adverse immune damage that affects human health [7]. Interferon-
 gamma (IFN-γ) is known as a glycoprotein whose molecular weight ranges between 17-25
kDa [8]. It was first distinguished in 1965 as a complete unit in Murine kupffer cells and
macrophages. Called IFN-inducing factor (IGIF) [9], also known as Type II interferon or
macrophage-activating factor (MAF). IFN-γ was act to support the immune system to
accomplish the cytolysis of target cells [10].

It stimulates the production of IFN-γ by active Th1 cells, CD4 + cells and CD8 + cells,
and can also be produced by NK cells and cytotoxic T cells (CTLs), and it can stimulate its
production by IL-12 and IL-18. [11], IFN directly inhibits virus replication and antigen
scaling [12]. IFN-γ belongs to the family of cytokines and the IFN-γ protein is produced by
the .IFNG gene initially. Interferon (IFNs) has been discovered as an anti-virus molecule and
act to inhibit a wide variety of viruses. Interferon are classified into two types, the first
includes IFN-α, IFN-, IFN-and the second type includes IFN-γ, which is a cytokine necessary
for innate and acquired immunity against viral infections and some bacterial and primary
infections, where it is considered an important activator for macrophages and a catalyst for
the expression of the histocompatibility complex molecule. Major Category II (MHC). IFN-γ
derivation is associated with a number of auto-inflammatory and autoimmune diseases, and
IFN-γ importance in the immune system stems in part from its ability to directly inhibit virus
replication, as IFN-γ is mostly produced by natural killer cells (NK) and natural killer cells
(NKT) as part of The innate immune response, and through the cytotoxic CD4 Th1 and CD8
T lymphocytes (CTL), once the specific immunity to the antigen develops as part of the
acquired immune response [13]. A gene in SARS-CoV-2 called a gene (orf3b), which acts on
the expression of a special protein that inhibits IFN expression and enhances the
pathogenicity of the virus [14].

The IFN-γ gene is located on the long arm of chromosome 12 at site 12q15 [15] It is
believed, through the study of the phenotypic polymorphism, that the IFN-gene has the ability
to influence some diseases, Immune diseases [16]. The study indicates that the A allele of the
IFN-γ + 874 gene has been significantly associated with Covid-19 infection as it indicates a
genetic risk factor associated with COVID-19. The study also shows that increasing IFN-γ
production during infection can prevent the recurrence of the Coronavirus and reduce the risk
of infection [17, 18]. This depends on the statistical analysis of PCR product evidence. The
aim of this study is to detect the phenotypic polymorphism of the IFN-γ T/A +874 gene by
using the replication impedance system with ARMS-PCR technology after isolating the DNA
from the studied samples to find out the relationship and linkage of the IFN-gene genotypes
with Covid 19.

2.Materials and methods

Blood samples collection from patients with type 2 diabetes

50 blood samples were collected from people with COVID whose ages ranged between
20-75 years and 30 samples from healthy subjects (standard group) whose ages ranged
between 19-63 years. Samples were collected from the Anbar Health Department / Public
Health Department / Public Health Laboratory in Anbar - Iraq, and 30 samples were collected
from healthy people (standard group) within the period from the first day of October 2020
until the end of November of the same year. The blood was collected in special tubes of 2.5
ml containing EDTA as an anticoagulant, which was used in the extraction of DNA. A
questionnaire form was organized in which the patient and healthy information were collected.

**DNA isolation and extraction kit**

gSTNCTM DNA Extraction Kit was used for DNA isolation that manufactured by the American company Geneaid. The concentration and purity of the DNA was determined by using a Nanodrop, and the purity was between 1.8 ± 1.5. The samples were then preserved at -20 °C until use.

**PCR Premix polymerase chain reaction kit**

An AccuPower® PCR PreMix kit was used in ARMS-PCR technology experiments, according to the accompanying leaflet by the Korean company BIONEER.

**Primers**

Three of the special primers were used to detect the IFN-γ T / A +874 mutant gene according to [19], as for the nucleotide sequences shown in Table (1) and prepared according to company procedure.

**DNA molecular weight markers**

The Molecular marker (US Company Promega) was used with a molecular weight 1.5 kg (2000 base pairs) and 100 base pairs.

**Detection of the IFN-γ T/A +874 gene**

The polymerase chain reaction method of the ARMS-PCR replication system was used to detect the IFN-γ +874 gene, and the master mix was prepared for the primers of the study according to the method [19], with some changes. An AccuPower® PCR PreMix kit was used in ARMS-PCR technology experiments, according to procedure of Korean company BIONEER. Specific A and Antisense primer were used to detect the A allele, Specific T and the Antisense primer to detect the T allele of the IFN-γ +874. The samples were placed in a thermo-cycler to amplifying DNA and adjusting the instrument program to obtain optimum conditions for interact. The first stage is the first denature template at a temperature of 95 °C and continues for 3 minutes. The first initial denaturation is set at 95 °C for 15 seconds and the first initial annealing is set at a temperature of 65 °C for a period of 50 seconds. The first extension is set at a temperature. 72 °C for a period of 40 seconds and the phases of the initial deformation, initiation and elongation of the first are controlled in 10 cycles. The second initial denaturation was set at a temperature of 95 °C for a period of 50 seconds, and the second annealing, set at a temperature of 55 °C for a period of 50 seconds, and the second extension was set at a temperature of 72 °C for a period of 50 seconds. Final extension was adjusted at 72 °C for a period of 7 minutes.

ARMS-PCR products of DNA of Covid-19 patients were loaded into the acarose gel device at a concentration of 1.5%. Then the molecular marker was carried genotype according to the accompanying procedure, which was about 1.5 kilo-base in size. The bromophenol blue loading dye for electrophoresis was added to each sample with a size of 3 μl to 7 μl for each sample in the electrophoresis. The samples were electrically migrated under 75 volts for 2-3 hours. staining the acarose gel with ethidium bromide (EtBr) stain for 20 minutes (the tincture
of ethidium bromide was prepared from the concentrated dye at a concentration of 10 mg/mL and prepared by dissolving it in distilled water for the purpose of obtaining a concentration of 0.5 μg/ml. The genotypes were observed and the enlarged pieces of DNA were photographed with a Gel documentation system equipped with a camera.

**Statistical analyzes**

The polymerase chain reaction product data were analyzed by using the Statistical Package for Social Sciences (SPSS), and the significant differences between the averages were compared by using Fisher's at a probability level P <0.05. Allele frequencies, genotypes, odds ratio (OR), and confidence intervals (CI) were analyzed by using the Compare 2 Ver.3.04 program manufactured by J. H. Abramson in 2003-2017 [20]. Results were analyzed by using the Hardy-Weinberg equilibrium law, according to the website www.had2know.com.

3. Results and Discussion

**COVID-19 samples and control sample**

The study included 50 blood samples for people with COVID-19, and the study included 30 blood samples for apparently healthy people that were counted as control samples. A genetic study was conducted on blood samples taken from patients with COVID-19 and healthy people (control sample).

**IFN-γ gene polymorphism**

IFN-γ genetic polymorphism was studied in patients with COVID-19 by using ARMS-PCR technology and compared with healthy samples (control samples). The results of the electrophoresis of the IFN-T / A +874 mutant gene amplified by ARMS-PCR showed the presence of two alleles, t and a, and through them, three genotypes were identified: TT, AT and AA in the sample of those infected with the Coronavirus and control sample, and when one package appeared in the t field and its absence in domain a, then the genotype is TT, and in the event that a beam appears in the domain a and does not appear in the t domain, then the genotype is AA, and when two packages appear in both domains t and a, then the genotype is AT, and as in Figures (1 & 2 respectively). The results of the repetitive distribution of the two alleles t and a of the mutant IFN-γ T / A +874 showed different results between patients with COVID-19 and healthy people, the t allele in the sample of patients with COVID-19 was (42%) compared to With the allele a, which is (58%), while the allele t was (71.67%), compared to the allele a, which is (28.33%), Figure (3).

Table (2) also shows that the frequency distribution show significant difference between patients with COVID-19 and healthy people, the a allele showed significant frequency in COVID-19 patients with a higher rate than the control sample by using Fisher's test and at a probability level P <0.05 and its value was (* 0.000). Odds Ratio (OR) was (3.49) with a confidence interval (CI). Confidence Intervals under the 95% value that ranged between (1.76-6.91), and the a allele appeared as a causative allele associated with the risk of infection with Covid-19 Etiological Fraction (EF). Where the value of the disease-causing fraction was (41.4%), while the t allele was shown as a preventive allele against COVID-19 infection, Preventive fraction (PF), and its value was as a preventive allele of disease (51.1%). Where it showed significant frequency in control sample with higher rates than the COVID-19 patients.
by using Fisher’s test, and the critical ratio was (0.39) with the Confidence Intervals under 95%, a value that ranged between (0.57-0.14).

The current study showed that the proportion of allele a was higher in patients when compared with the standard sample, while the t allele showed a higher percentage in the standard sample of patients. These results is in agree with (19), who referred that the percentage of allele a was higher in patients compared to the standard sample, while the t allele showed a higher percentage in the standard sample. The high frequency of the a allele, in a significant way among the infected, indicates the great role that this allele plays with the risk of Corona patients, while the low frequency of the t allele in the infected and its elevation in the standard sample shows the importance of this allele as a preventive factor from the risk of infection with this disease.

The results of the statistical and genetic analysis of ARMS-PCR technology for the mutated gene IFN-γ T / A +874, using the Hardy-Weinberg equilibrium law, showed three genotypes in the sample of those infected with Coronavirus and the control sample which is TT, AT and AA. The results showed a variation in the frequency of genotypes between patients with COVID-19 and healthy people, as the AA genotype showed a higher percentage in the patient sample compared to the control sample and the proportions were (40%) and (16.67%), respectively, and there was a significant difference of Its value (*) at a P <0.05 level using Fisher's test. The value of Odds Ratio was (3.33) and the duration of confidence was between (9.99-1.11), and the AA genotype appeared as a genotype associated with the risk of infection with the Coronavirus, as the value of the risk causing the disease was (28%). The TT genotype showed a higher percentage in the healthy sample compared to the patient sample, and the ratios were (60%) (24%), respectively, the TT genotype was shown as a genotype significantly associated with the preventive aspect of the risk of developing COVID-19 preventive fraction (PF). Its protective value (47.4%) showed significant frequency in the control sample with higher rates than the infected sample, and the statistical analysis when using Fisher's test showed that the TT genotype was significantly different among the infected sample compared with the control sample and at a probability level P <0.05 and its value was (* 0.001), and odds ratio was (0.21), with the Confidence Intervals duration under the 95% ratio, and the value ranged between (0.55-0.08).

The AT genotype also appeared as a genotype associated with the side that causes the risk of infection with COVID-19 and its value was as a pathogen (16.5%), but in a non-significant form, as no significant difference between the infected sample and the standard sample was seen at a probability level P <0.05, its value was 0.176. The genotype AT showed a higher percentage of infected people compared to the control sample, and the percentages were (36%) and (23.33%), respectively, the value of the critical ratio was (1.85) and the confidence interval was between (0.76-5.07), as in Figure (4) Table (3).

It appears that the two heterozygous AA and TA genotypes are associated with the risk and development of infection with COVID-19, but the association of the AA genotype with the pathological side of the risk of infection with COVID-19 was high, and it was found that the genotype TT appeared as a genotype associated with the preventive side of the risk of infection with COVID-19, and it can be adopted as a preventive indicator of the risk of developing the disease, and the results also show that the homozygote AA genotype and the heterozygote TA are more dangerous in patients with COVID-19, and these results indicate the importance of the role of the TT genotype as a protective genotype of Infection with
COVID-19. These results have been confirmed by many studies that show that the A allele may be a reliable indicator in diagnosing the risk of developing the disease and stopping beta cells from producing, and it was suggested that the T allele polymorphism and the TT genotype do not have a significant role with the development of SARS, Rather, it could be a preventive indicator of the risk of developing the disease, in contrast to the A allele and the AA and AT genotypes, which show a significant association with the risk of developing SARS [19].

A study showed that increased expression of the IFN-gene plays a major role in the development of some diseases [21]. If IFN-γ is stops working, or by blocking the expression of the IFN-gene and disrupting the IFN-receptor, it increases disease risk [22]. The results of a study indicated strong associations of IFNγ-874A / T with viral infections. It has been suggested that individuals with the homozygous IFNγ-874 A / A genotype had a significantly higher risk of different viral infections [23]. Also, in Korean patients, there was a higher frequency of the T allele for the IFNγ gene in healthy subjects compared to the affected group [24].

A study showed that increased expression of the IFN-gene plays a major role in the development of some diseases [21]. If IFN-γ is stops working, or by blocking the expression of the IFN-gene and disrupting the IFN-γ receptor, it increases disease risk [22]. The results of a study indicated strong associations of IFNγ-874A / T with viral infections. It has been suggested that individuals with the homozygous IFNγ-874 A / A genotype had a significantly higher risk of different viral infections [23]. Also, in Korean patients, there was a higher frequency of the T allele for the IFNγ gene in healthy group compared to the infected group [24].

The COVID-19 virus, its degree of severity, may be associated with the regulation of IFN-γ production, and this regulation depends on the expression or inhibition of IFN-γ (25). A decrease in IFN-γ production could weaken its response to viral infections including COVID-19, making people more vulnerable to infection with this virus. The study indicates that the IFN-γ + 874A allele has been significantly associated with infection with the COVID-19 virus as it indicates a genetic risk factor associated with SARS. The study also shows that increasing IFN-γ production during infection can prevent the recurrence of the COVID-19 virus and reduce the risk of infection [17, 18]. IFNγ binding on cells through its own receptor leads to STAT-1 activation, which in turn binds to transcription factors that will stimulate MHC-II and protein kinase (PKR) and increase ribonuclease L expression that works on resistance to the virus, thus inactivating or failing. The IFNγ signaling pathway gives a higher susceptibility to virus infection, indicating that IFNγ secretion may be a major function of suppressing various viral infections [26].

**Table (1): Sequences of primers for IFN-γ T / A +874 gene**

| Gene Locus | Sequence of Generic primer (antisense) | Sequence of Sense primers |
|------------|--------------------------------------|---------------------------|

6
Table (2): Repeats of the two alleles t and a of the IFN-γ A/T +874 mutant gene in infected sample and control sample

| Gene     | Allele | Infected samples (100) | Control samples (60) | (95% CI) OR | P value |
|----------|--------|------------------------|----------------------|-------------|---------|
| IFN-γ A/T +874 | t      | 42(42%)                | 43(71.67%)           | 0.39(0.57-0.14) | *0.000   |
|          | P.F    | 51.1%                  |                      |             |         |
|          | a      | 58(58%)                | 17(28.33%)           | 3.49(1.76-6.91) |         |
|          | E.F    | 41.4%                  |                      |             |         |

OR = Odds ratio, CI = Confidence Intervals, PF = Preventive faction, EF = Etiological faction, * = significant difference at P <0.05. Fisher’s test.

Table (3): The frequencies of the genotypes for IFN-γ A/T +874 mutant gene in infected sample and control sample

| Gene     | Genotype | Infected samples (100) | Control samples (60) | (95% CI) OR | P value |
|----------|----------|------------------------|----------------------|-------------|---------|
| IFN-γ A/T +874 | TT      | 12(24%)                | 18(60%)              | 0.21(0.08-0.55) | *0.001   |
|          | P.F      | 47.4%                  |                      |             |         |
|          | AT       | 18(36%)                | 7(23.33%)            | 1.85(0.76-5.07) | 0.176    |
|          | E.F      | 16.5%                  |                      |             |         |
|          | AA       | 20(20%)                | 5(16.67%)            | 3.33(9.99-1.11) | *0.025   |
| OR = Odds ratio, CI = Confidence Intervals, PF = Preventive faction, EF = Etiological faction, * = significant difference at P <0.05. Fisher's test. |

**Figure (1):** The electrophoresis of the IFN-gene to the T / A + 874 mutation site showing the two alleles t and a in infected samples.

![Figure 1](image1)

**Figure (2):** The repeat ratio of the two alleles t and a seen for the IFN-γ A / T gene +874

![Figure 2](image2)
Figure (3): shows the genotypes seen for the IFN-γ T/A +874 mutant gene in infected and control samples.

4. References

[1] Schalk, A. & Hawn, M. C. (1931). An apparently new respiratory disease of baby chicks. J. Am. Vet. Med. Assoc. 78: 413–423
[2] Lai M.M. and Cavanagh D. (1997). The molecular biology of coronaviruses. Adv. Vir. Res.48:1–100.
[3] Qing Ye, Bili Wang, Jianhua Mao . The pathogenesis and treatment of the `Cytokine Storm' in COVID-19, 2020.
[4] Lai C.; C.Shih T.; P.Ko W.-C.Tang H.-J.Hsueh P. R. (2020). Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19): The epidemic and the challenges.Int J Antimicrob Agents.
[5] N. Chen, M. Zhou, X. Dong, et al./Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study Lancet., 395 (2020), pp. 507-513.
[6] Chu, Daniel KW, et al. "Molecular diagnosis of a novel coronavirus (2019-nCoV) causing an outbreak of pneumonia." Clinical chemistry 66.4 (2020): 549-555
[7] Channappanavar R.Fehr A.R.Vijay R.Mack M. Zhao J.Meyerholzd.K.et.al.Dysregulated Type I Interferon and inflammatory monocyte-macrophage responses cause lethal pneumonia in SARS-CoV-Infected Mice.Cell Host Microbe. 2016; 19 (PubMed PMID: 26867177. eng): 181-193
[8] Curfs, J., Meis, J. F. and Korstanje, J. A. (1997). A primer on cytokines: Sources, receptors, effects, and inducers. Clini. Microbiol. Rev. 10(2):742-780
[9] Feghali, C. A. and Wright, M. (1997). Cytokines in acute and chronic inflammation. Frontiers in Biosci. J. 2(1):12-26.
[10] Stalenhoef, J. E., Alisjahbana, B., Nelwan, E. J., van der Ven-Jongekrijg, J., Ottenhoff, T. H., van der Meer, J. W., Nelwan, R. H., Netea, M. G. and van Crevel, R. (2008). The role of in-terferon-gamma in the increased tuberculosis risk in type 2 dia-betes mellitus. Eur. J. Clin. Microbiol. Infect Dis. 27:97-103.
[11] Schroder, K., Hertzig, P., Ravasi, T. and Hume, D. (2004). Interferon-y: an overview of signals, mechanism and functions. J. Leukocyte Biol. 75:163-189.
[12] Saha, B.; Jyothis Prasanna, S.; Chandrasekar, B. and Nandi, D. (2010). Gene modulation and immunoregulatory roles of interferon gamma. Cytokine. 50:1–14.

[13] Trinchieri, G. (2010). Type I interferon: friend or for published. J. Exp. Medici. 207(10):2053-2063.

[14] Chan, JF.; Kok, KH.; Zhu, Z.; Chu, KK.; Yuan, TS. and Yuen, KY. (2020). Genomic characterization of the 2019 novel human-pathogenic coronavirus isolated from a patient with atypical pneumonia after visiting Wuhan. Emerg Microb Infect. 9:221–36.

[15] Hardy, M. P.; Owczarek, C.; Jermiin, L.; Eijdeback, M. and Hertzog, P. (2004). Characterization of the type I interferon locus and identification of novel genes. J. Genomics. 84(2):331-345.

[16] Cantor, M. J.; Nickerson, P. and Bernstein, C. N. (2005). The role of cytokine gene polymorphisms in determining disease susceptibility and phenotype in inflammatory bowel disease. Am. J. Gastroenterol.100:1134–1142.

[17] Scagnolari C.; Vicenzi E.; Bellomini F.; Stillitano MG.; Pinna D.; Poli G.; Clementi M.; Dianzani F. and Antonelli G. (2020). Increased sensitivity of SARS-coronavirus to a combination of human type I and type II interferons. Antivir Ther. 9:1003-11.

[18] Sainz B.; Mossel EC.; Peters CJ. and Garry RF. (2020). Interferon-beta and interferon-gamma synergistically inhibit the replication of severe acute respiratory syndrome-associated coronavirus (SARS-CoV). Vir. 329:11-7.

[19] Chong, W.P., Ip, W.E., Tso, G.H.W. et al. The interferon gamma gene polymorphism +874 A/T is associated with severe acute respiratory syndrome. BMC Infect Dis 6, 82 (2006). https://doi.org/10.1186/1471-2334-6-82.

[20] Dhakaan A. A.(2017). The Allelic and Polymorphism Association of Tumor Necrosis Factor-alpha Gene (-308 G/A Genotype) in Some Iraqi Rheumatoid Arthritis Patients. International Journal of Sciences: Basic and Applied Research (IJSBAR) .36( 5): 302-309.

[21] Cope, A. P.; Liblau, R. S.; Yang, X. D.; Congia, M.; Schreiber, R. D.; Probert, L.; Kollias, G. and Mcdevitt, H. O. (1997). Chronic tumor necrosis factor alters T cell responses by attenuating T cell receptor signaling. J. Exp. Med. 185:1573–1584.

[22] Akalin, E. and Murphy, B.(2001). Gene polymorphisms and transplantation.J. Curr. Opin. Immununol. 13(5):572-6.

[23] Sobti R. C.; Salih A. M. and B. Nega (2010). Insights into the role of IL-12B and IFN-γ cytokine gene polymorphisms in HIV-1/ AIDS infection. J. Folia Bio. 56(3): 110–115.

[24] Kang, M.W.; Pyo C.-W. and Wie S.-H. (2006). Associations of IFN-γ polymorphism with HIV-1 infection in the Korean population,” AIDS Research and Human Retroviruses, vol. 22, no. 3, pp. 297– 299.

[25] Pravica V.; Perrey C.; Stevens A.; Lee JH. and Hutchinson IV. (2000). A single nucleotide polymorphism in the first intron of the human IFN-gamma gene: absolute correlation with a polymorphic CA microsatellite marker of high IFN-gamma production. Hum Immunol 61:863-866.

[26] Chan L. L. Y.; Cheung B. K. W.; Li J. C. B. and Lau A. S. Y. (2010). A role for STAT3 and cathepsin S in IL-10 down-regulation of IFN-γ-induced MHC class II molecule on primary human blood macrophages. J. Leukocyte Bio. 88(2): 303–311.