OXIDATIVE STRESS MEASUREMENT AND EVALUATION GUIDE

RELASSAY DIAGNOSTICS
2012

OXIDATIVE STRESS, MEASUREMENT AND EVALUATION

Free radicals are metabolic by products which are formed during energy conversion of nutrients in our bodies by using oxygen. Free radicals (reactive oxygen species) are in unstable form and in order to become stable they attack and damage cells. Antioxidants neutralize free radicals to protect cells from damage. Free radicals and antioxidants must be in balance thus antioxidants neutralize free radicals. If free radical level increased against antioxidant level oxidative damage occurs and this state is called oxidative stress.

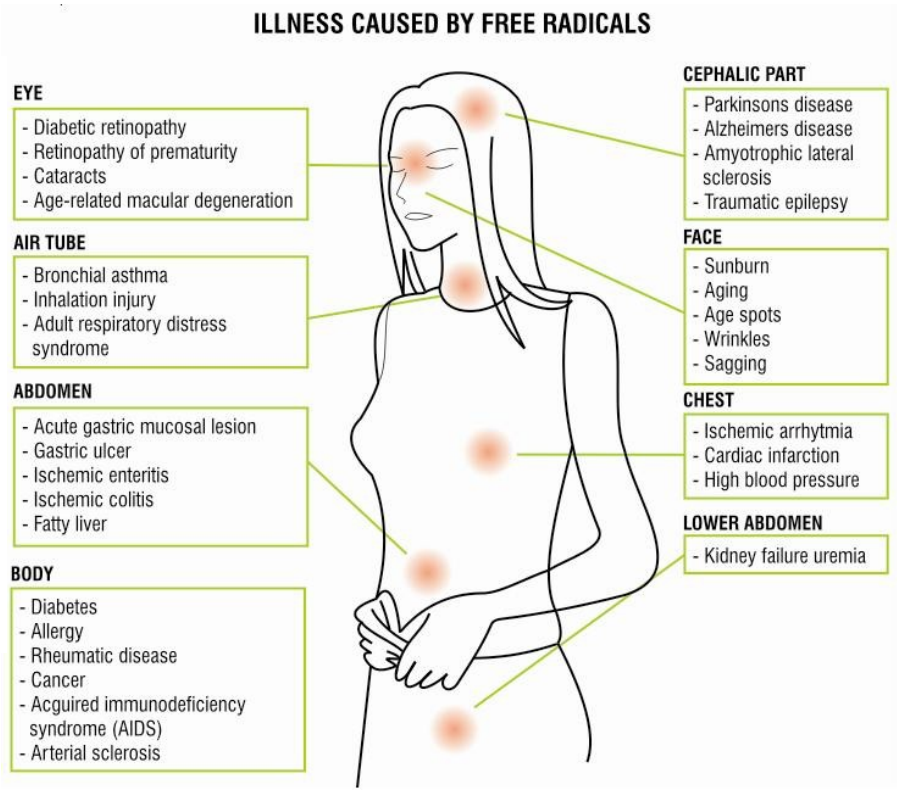
Oxidative stress is not a disease, but it is a factor that can cause or acelerate the disease. Genarally it is an important stimulus for preventive health measures. But the dangerous is oxidative stress is absence of any symptoms. If this condition is not diagnosted and treated, could result serious health problems and diseases. Exposure to toxins or pathogens, poor antioxidant defense system, irregular life style, extreme intense exercise and daily metabolic products, causes oxidative stress.

In some cases oxidative stress increase higher than normal levels.

For example;

- Pregnancy
- Hormone replacement therapy
- Birth control pills
- After heavy exercise
- Excessive exposure to sunlight
- Excessive alcohol consumption
- Electromagnetic radiation
- Smoking
- Air Pollution
- Malnutrition
- Chronic inflammation

In such cases, must pay more attention to the antioxidant intake.



Reducing free radicals and increasing antioxidants by diet and supplements

There are many factors affecting the antioxidant defense system of the body. Individual genetic structure and environmental factors that body exposed. Unfortunately, the modern way of life, environmental pollution, stress, low-quality foods, unbalanced nutrition, causes remain to expose to more free radicals. So physilogically produced antioxidants are not enough to neutralize free radicals.

By rich in antioxidants diet defense of the body against free radicals can be increased.

Diet contains fruits, vegetables and oily seeds vitamins, antioxidants and other antioxidant micronutrients creates a good source of exogenous character and increases the cellular response to oxidative stress.

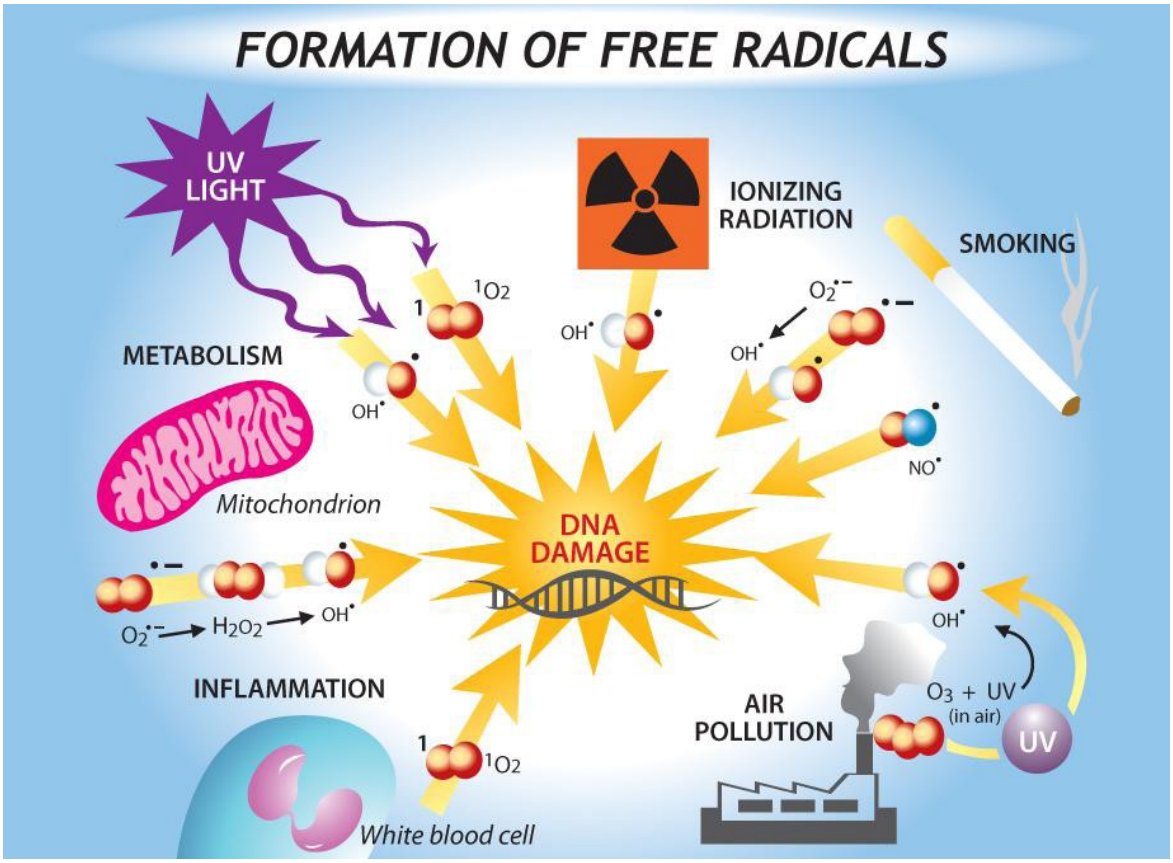
For example, essential vitamins such as vitamin C has suppressive and reducer effect on many different reactive oxygen species and not synthesized by human body. Epidemiological studies support that people who eat rich in antioxidants healths are beter. The World Health Organization (WHO) recommends as a result of epidemiological studies, daily consumption of at least 400g of fruit and vegetables.

Every part of the cell is maintained by a different antioxidants. Therefore fed on by a variety of nutrition is important. For example some of the food items protects against free radicals inside the cell, outside the cell wall, inside the blood surrounding the cell and the cell membrane. For this reason, we need both exogenous and endogenous antioxidants

As examples of antioxidants:

- Vitamin C, protects vitamin E and refreshes the extracellular fluid.
- Vitamin E protects cell wall.
- Carotenoids (In the content of the fruits and vegetables that people consumed, founded over 60 caretenoids, beta-carotenoids, laykopen, lutein and other colored compounds, yellow, red, orange.)
- Thiols protects inside of the cell (sulfur-containing compounds such as lipoic acid, and glutathione) .
- Co-enzyme Q10 protects mitochondria and slows aging.
- Flavonoids protects DNA,elastin and collagen tissue, thus slow down the aging process.
- Enzymes including superoxide dismutase, glutathione peroxidase, catalase, can neutralize free radicals in the cells. They can not gotten from the outside they are endogenous antioxidants produced by the body. However, they need the dietary selenium, copper, zinc and trace minerals such as manganese.

Making frequent changes in the diet may not enough to organize antioxidant intake. Diet should be prepared by an expert and necessary foods contains antioxidants should added to the diet.



- Get a balanced diet, consume adequate amounts of fruits and vegetables , if possible, be sure to choose organic of them. Drink plenty of water, avoid fatty, fried and processed foods.
- Avoid excessive consumption of sugar and salt, and choose products that are not refined.
- Limit your alcohol intake.
- Consume more tea (green tea), and less coffee.
- Do mid-level exercise on a regular basis, but do not overdo it. Much forcing your body, increase the level of free radicals.
- Stay as far away as possible from pollution, toxic materials causes oxidative stress. Stay away from industrial pollution, car pollution, cigarette smoke, and toxic substances in food.
- Insomnia causes oxidative stress, thus try to get regular sleep.
- Avoid long-term exposure to ultra-violet rays and being under the sun. Apply strong protection creams under the sun.
- Regularly measure your blood sugar, cholesterol and blood pressure and keep under control.
- If you are taking hormones (birth control pills or hormone replacement therapy) keep control of your oxidative stress.
- Keep away from excessive mental and physical stress.
- Give yourself time to relax and do things that make you happy.

OXIDATIVE STRESS MEASUREMENT

Serum (or plasma) concentrations of different antioxidants can be measured in laboratories separately, but the measurements are time-consuming, labor-intensive, costly, and require complicated techniques. Because the measurement measurement of different antioxidant molecules separately is not practical and their antioxidant effects are not additive.

With Rel Assay Total Antioxidant Status(TAS) and Total Oxidant Status (TOS) kits you can easily measure non-enzymatic all antioxidant and oxidant molecules.

For measurement procedures and booklets you can visit www.relassay.com, send e-mail to relassay@relassay.com or make a phone call to +90 342 220 49 87.

Collection of samples

For this measurement serum is used as sample. Hemolyzed or lipemic serum is not required. The sample should be taken after fasting for 10-12 hours. Sample must not taken after exercise.

Storage of samples

Serum 2-8 ◦ C 24 hours
Serum -20 ◦ C 3 months
Serum -80 ◦ C 1 Year

TAS Test Principle

Antioxidants in the sample reduce dark blue-green colored ABTS radical to colorless reduced ABTS form. The change of absorbance at 660 nm is related with total antioxidant level of the sample. The assay is calibrated with a stable antioxidant standard solution which is traditionally named as Trolox Equivalent that is a vitamin E analog .

TOS Test Principle

Oxidants present in the sample oxidize the ferrous ion–chelator complex to ferric ion. The oxidation reaction is prolonged by enhancer molecules, which are abundantly present in the reaction medium. The ferric ion makes a colored complex with chromogen in an acidic medium. The color intensity, which can be measured spectrophotometrically, is related to the total amount of oxidant molecules present in the sample. The assay is calibrated with hydrogen peroxide and the results are expressed in terms of micromolar hydrogen peroxide equivalent per liter (µmol H2O2Equiv./L)

EVALUATİON OF THE RESULTS

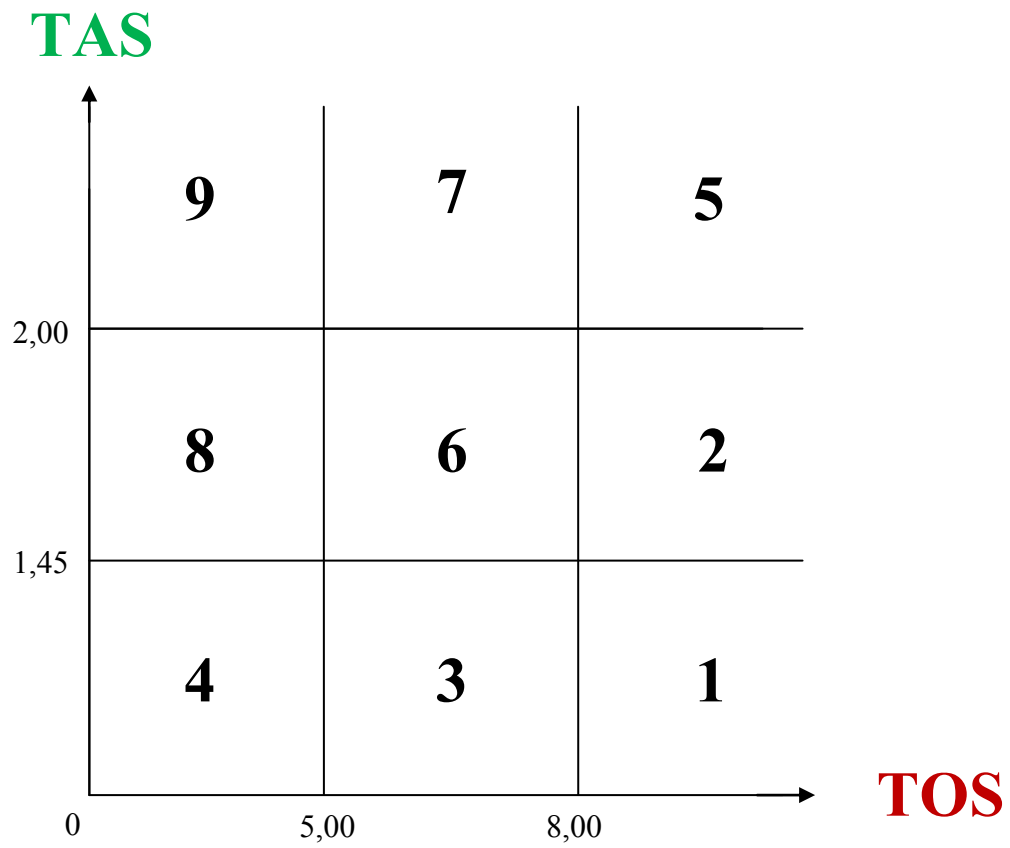
Rel Assay TAS and TOS test results can be evaluated independently in accordance with the followingtables or we recommend to put the results of both to 9-diagnosquare to evaluate.

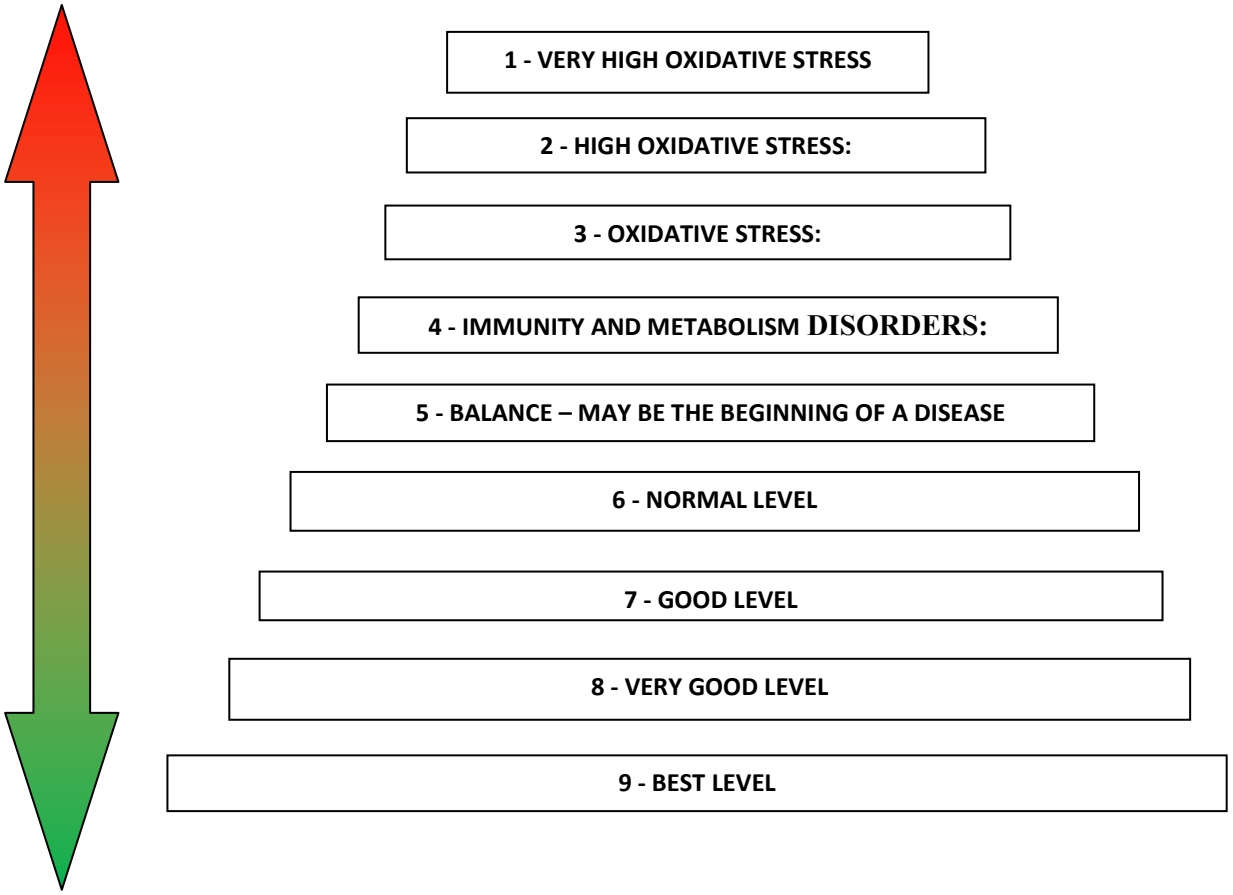
TAS REFERENCE VALUES (mmol Trolox Equiv./L)		
>2,0		Very good
1,45	2,00	Normal
1,20	1,45	Tolerable
1,00	1,20	Low Antioxidant Level
<1,20		Very Low Antioxidant Level

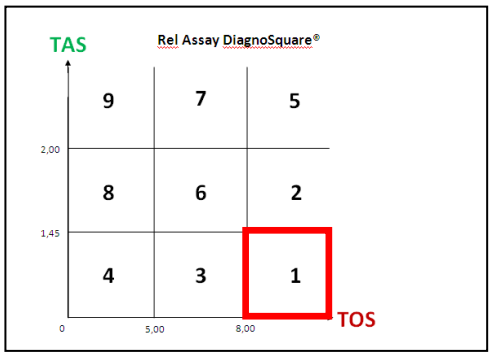
TOS REFERENCE VALUES (µmol H_2O_2 Equiv./L)		
<5,00		Very Good
8,00	5,00	Normal
12,00	8,00	High Oxidant Level
>12,00		Very High Oxidant Level

- This reference range is prepared in the light of the scientific data in accordance with the international literature.
- Samples used in this study carried out in Turkey by the Turkish population.
- In this study, samples were taken from 20-60 years of age healthy individuals has taken place as"normal" status.
- It is recommended to evaluate the results of TAS and TOS according to the 9-DiagnoSquare .
- It should be noted that each laboratory determine its own reference range. This study is a sample, prepared by the manufacturer by itself. The manufacturer does not accept responsibility for any problems arising from the use of this data.

Rel Assay DiagnoSquare®

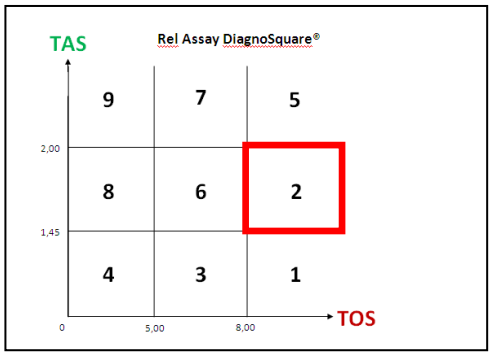






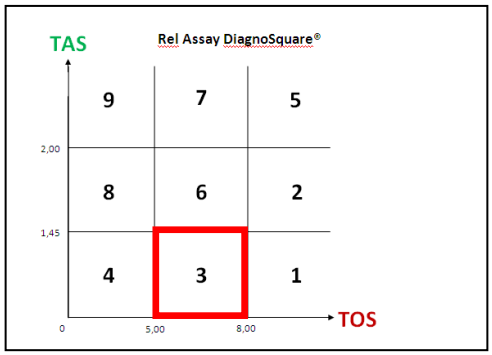
1 - VERY HIGH OXIDATIVE STRESS

Oxidants too much, antioxidations is very low, this is quite dangerous. Individuals in this region must be suspected from degenerative diseases, cardiovascular diseases and cancer diseases and advanced examinations must be consulted. In addition, use of medication, chemotherapy and radiation therapy can cause very high oxidative damage. The individual must be examined in detail, besise specifically treatment must be supported by antioxidants. TAS TOS levels must be checked every week.



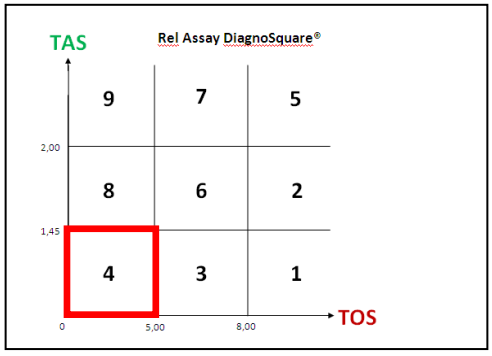
2 - HIGH OXIDATIVE STRESS

The state of high oxidant level beside normal antioxidant level. This may be initial phase of many degenerative diseases. Hormone replacement therapy, birth control pills, sedentary lifestyle, alcohol, smoking and even obesity can cause high-oxidative level. In addition it can be seen in pregnancy. In addition, high sporting activity can cause this case. Patient history must listened carefully and, if necessary supported by other tests. Oxidant reducer treatment must be executed and TAS TOS levels must be checked for every 15 days.



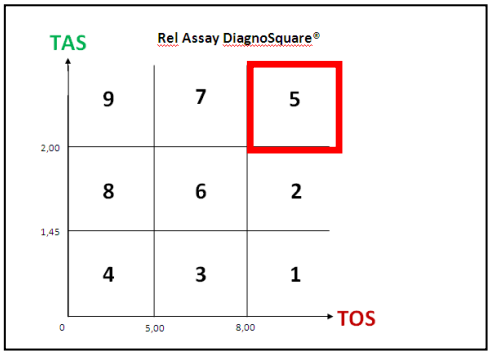
3 - OXIDATIVE STRESS

The condition with normal levels of oxidants, but with not enough antioxidants. In these individuals due to malnutrition, attention deficit, infectious diseases, the slow pace of growth and development in children, mental retardation, premature aging and such as diseases can be seen. Individuals lifestyles and eating habits could be examined in detail and they should supported with antioxidants by a dietitian who has results in this area. TAS and TOS tests should be repeated after 15 days.



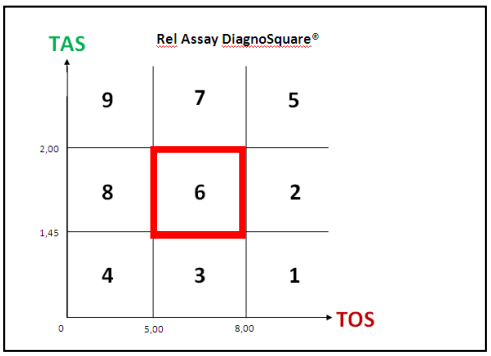
4 - IMMUNITY AND METABOLISM DISORDERS

Oxidants Antioxidants are both low than normal, this situation is rare. Decrease in the level of oxidants can interpreted good but decrease in antioxidants is not a good situation at all. The results can be like this at the breakdowns of immune system. Metabolic diseases should be considered in such situations, and further investigations should be requested. TAS and TOS tests should be repeated after 15 days.



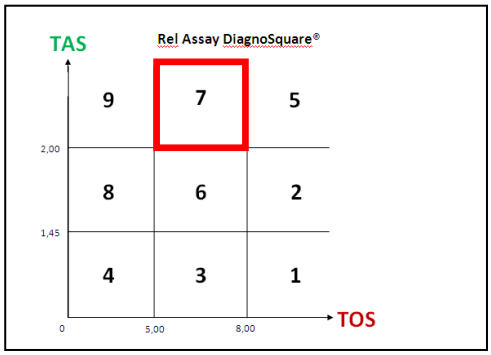
5 - BALANCE – MAY BE THE BEGINNING OF A DISEASE

Oxidants and antioxidants both high, this situation is rare. This occurs usually after intense exercise in athletes. This condition may be met normal for athelete but for healthy individuals increase of oxidants that much can cause some problems. In healthy individuals this can be observed at pre-disease cases or cases of acute exposure to poor environmental conditions. Required specific therapy must be imlemented and to confirm derease in oxidant level, test should be repeated after 15 days.



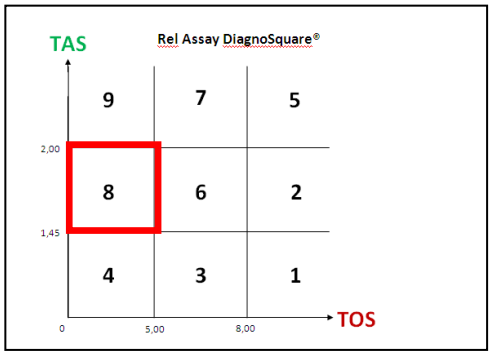
6 - NORMAL LEVEL

Previous studies have shown that, under normal conditions Oxidants and Antioxidants is in equilibrium for the organism, and the vast majority of healthy individuals are in this group. Nowadays, enough information is available about the importance of antioxidant defense system and the hazards of oxidants. Therefore should pay attention to lifstyle to not to slide this balance toward oxidative stress and test should be repeated every 2 months.



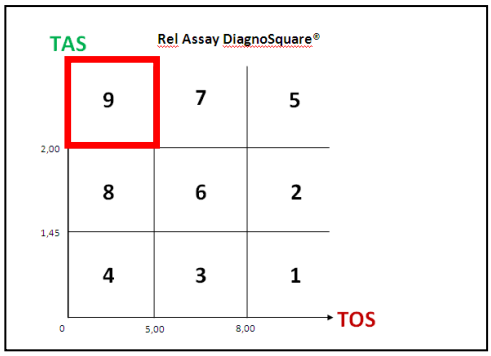
7 - GOOD LEVEL

The normal level of oxidants, and higher antioxidant level, in this situation there is no oxidative stress. Generally individuals who care about diversity of plant foods in their diet have this results. In addition, this situation can be found for the individuals who takes antioxidant support.



8 - VERY GOOD LEVEL

Oxidants is at the below of normal level, antioxidants is in normal level, in this situation there is no oxidative stress. Decrease in oxidants could be related to lifestyle and living environment. Generally, found in individuals who is far away from city life and naturally nourished.



9 - BEST LEVEL

Above normal level of antioxidants, below normal level of oxidants, this situation is described as the best level. If the individual is not taking antioxidant supplements and reached to this situation means individual is at disease risk-free group.

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