Food Oils and Fats: Chemistry and Technology

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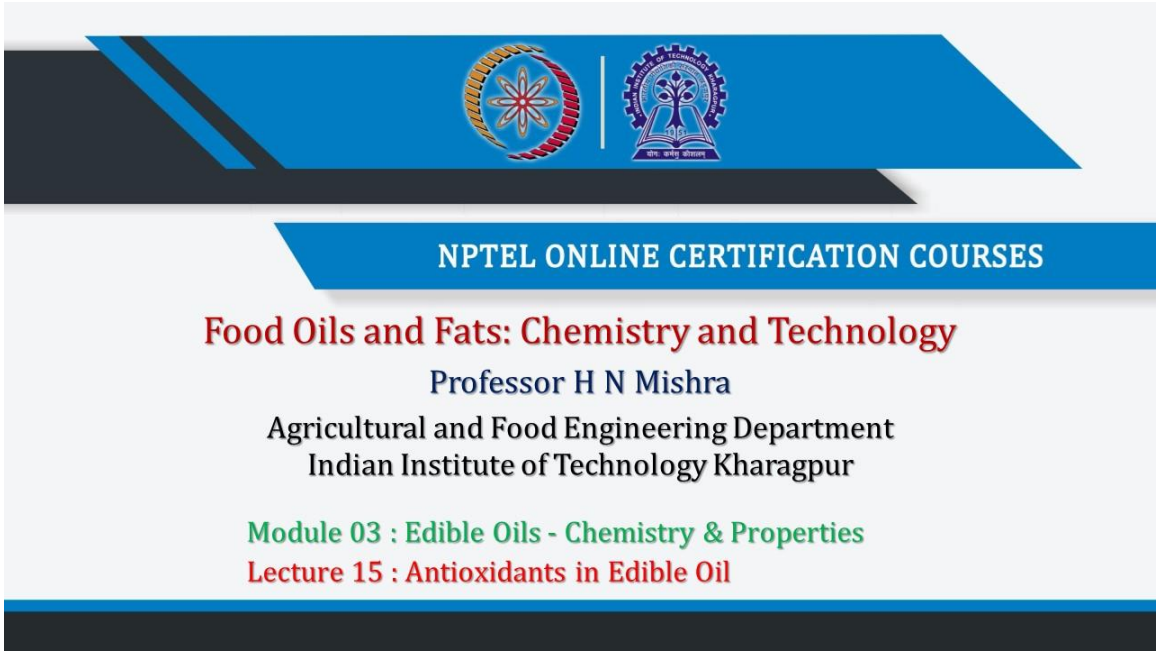
Module 03 : Edible Oils - Chemistry & Properties

Lecture 15: Antioxidants in Edible Oil



Hello, everybody. Namaskar. Now, we are in the 15th lecture of this course and in this lecture in the next half an hour, we will study about antioxidants, which are naturally present antioxidant as well as added antioxidants in edible oils. In the earlier class, the last class, we discussed about autoxidation process and while discussing the preventive measures or how these oxidation, autoxidation can be prevented, we elaborated different methods. And there one, important method was that use of antioxidants. So, what are antioxidants that I elaborated in the last class.

Concepts Covered

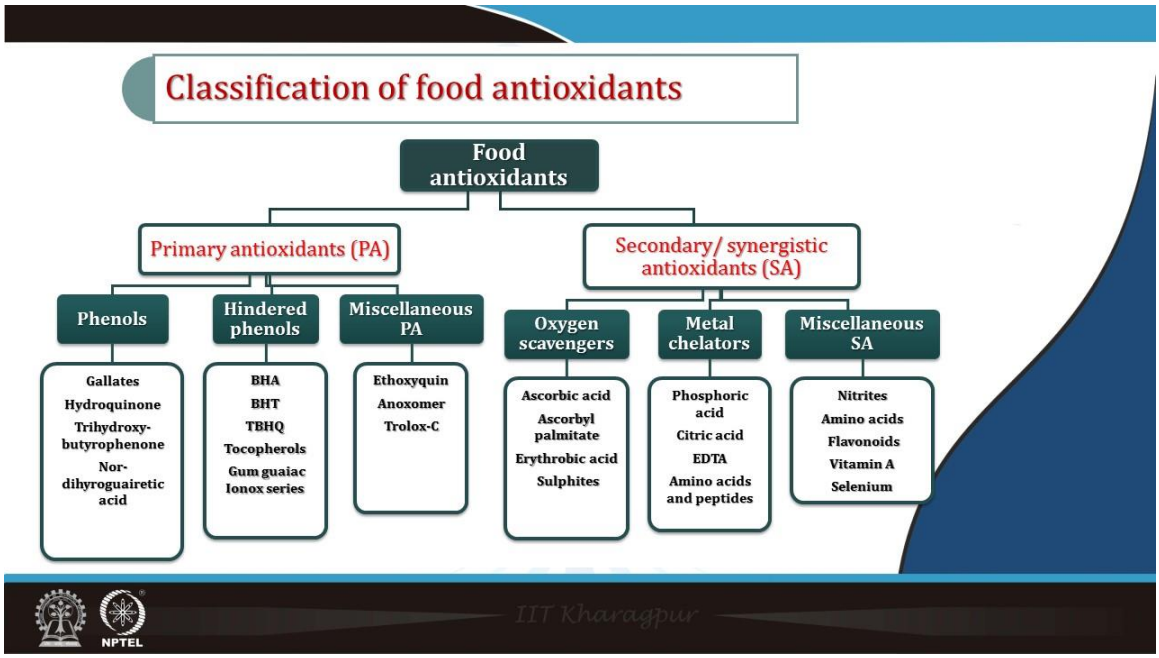
- Naturally present antioxidants
- Synthetic antioxidants
- Mechanism of action of antioxidants
- Total antioxidant capacity of vegetable oils - DPPH, FRAP
- Oxidative stability index

Today in this class, we will discuss about what are the naturally present antioxidants, what are synthetic antioxidants that are frequently used in edible oils and fats, mechanism of action of antioxidants, total antioxidant capacity of vegetable oil like DPPH, FRAP, etcetera and oxidative stability index, how they are determined and what does it mean.

Antioxidants

- Substances which inhibit or delay the oxidation of a substrate when present at concentrations lower than the oxidizing substrate.
- Are used to counteract the deterioration of stored food products.
- Function by interfering with the free-radical chain reactions.
- Reduce the primary radicals to non-radical species.

So, antioxidants. I earlier also told. Now, these are the substances which from the name itself you can take it out that the substances which inhibit or delay the oxidation of a substrate when present at a concentration lower than the oxidizing substrate, that is, they are active at a very low concentration. Antioxidants are used to counteract the deterioration of stored food products and in this case, particularly, we are talking about edible oils. So, these antioxidants, they are used in edible oil to prevent their oxidation, autoxidation process. Antioxidants function by interfering with the free radical chain reaction process. In the last class, we saw that how the autoxidation takes place; that is, the initiation reaction where the free radicals are formed; then propagation, that is the chain reaction and then finally, termination. So, it is basically, that is, once the antioxidants are formed, that is, the free radicals, sorry, once the free radicals are formed, that is, thereby initiation process, then immediately antioxidant become active and they interrupt the next process, that is, the propagation process though. So, they function by interfering with the free radical chain reaction and they reduce the primary radicals to non-radical species.

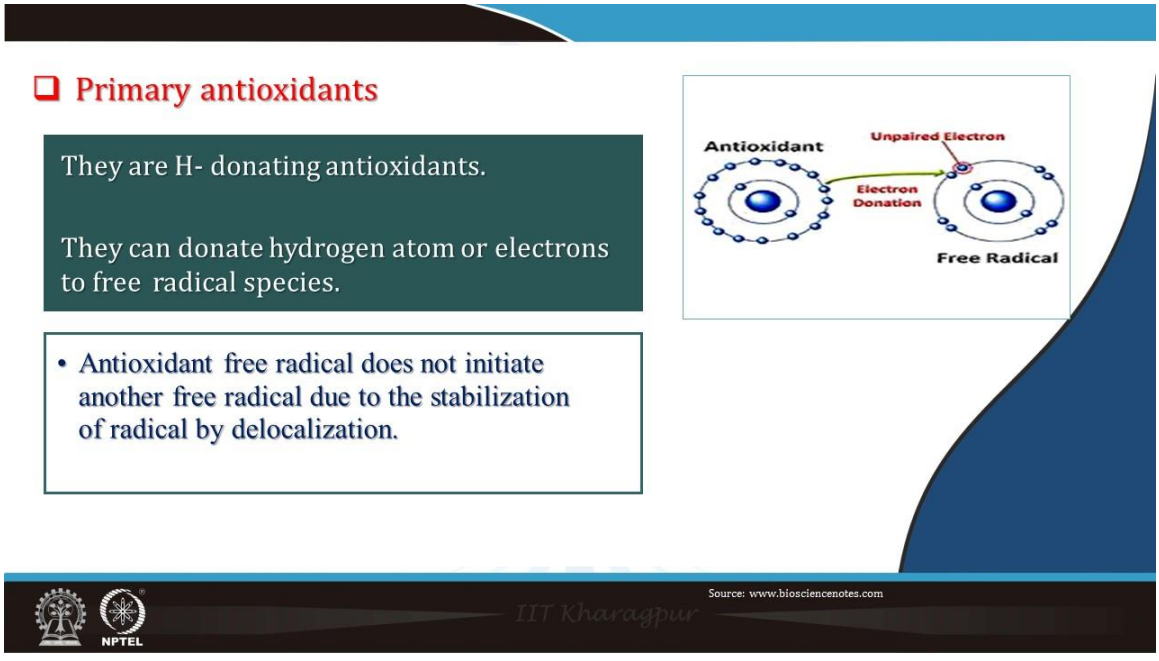


So, various food antioxidants are elaborated in this table and they can be broadly classified into two groups. One is the primary antioxidant and other is the secondary antioxidant or which are the which may also be called sometime as synergistic

antioxidants. So, the primary antioxidants includes phenols like gallates, hydroquinone, trihydroxybutyrophone, nordihyroyguairetic acid and so on.

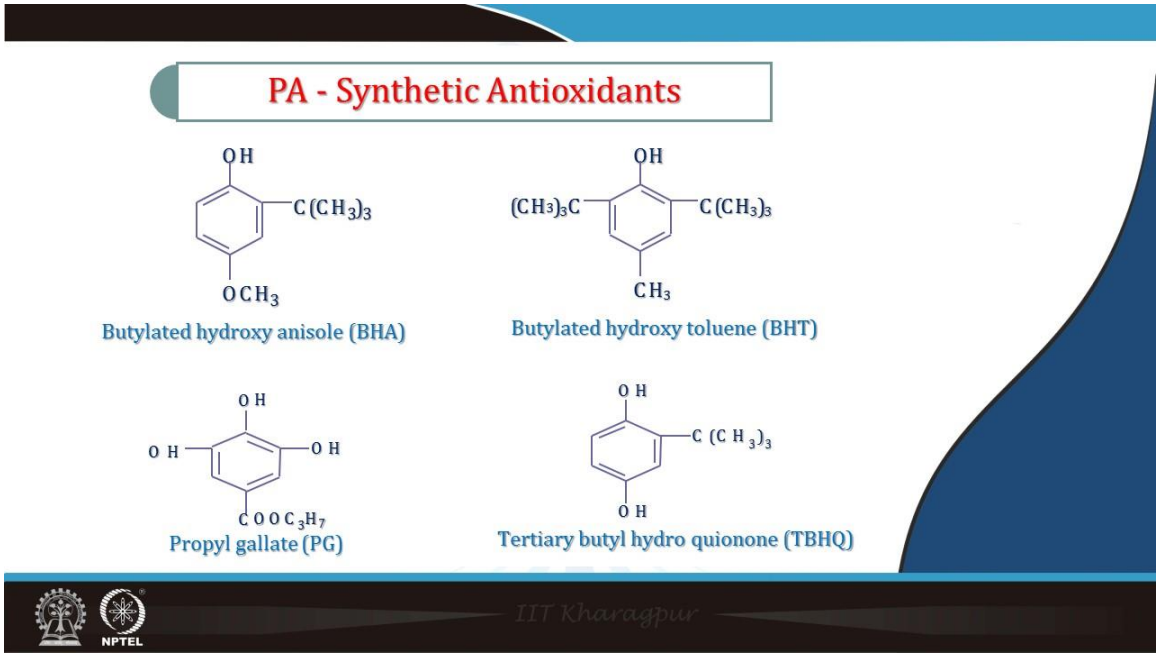
The hindered phenols they are also included in the primary antioxidants. And hindered phenols are. Examples of hindered phenols are BHA that is butylated hydroxy anisole, butylated hydroxy toluene, tertiary butyl hydroxyquinone, tocopherols, gum guaiac, ionox series and so on. Then there are other miscellaneous primary antioxidants like ethoxyquin, anoxomer and trolox-c. So, these are the primary antioxidants, ok. Then the secondary antioxidants, they, which work as a oxygen scavenger or metal chelators or other by miscellaneous. So, major oxygen scavenger, that is, what they do. They scavenge the oxygen molecules and make them unavailable for the oxidation reaction.

Then these prominent oxygen scavengers, which are used in edible oils and fats are ascorbic acid, ascorbyl palmitate, erythrobic acid, sulfites and so on. Then metal chelators are phosphoric acid, citric acid, EDTA, amino acids and peptides and the miscellaneous synergistic antioxidants are nitrites, amino acids, flavonoids, vitamin A, selenium, etcetera, ok.

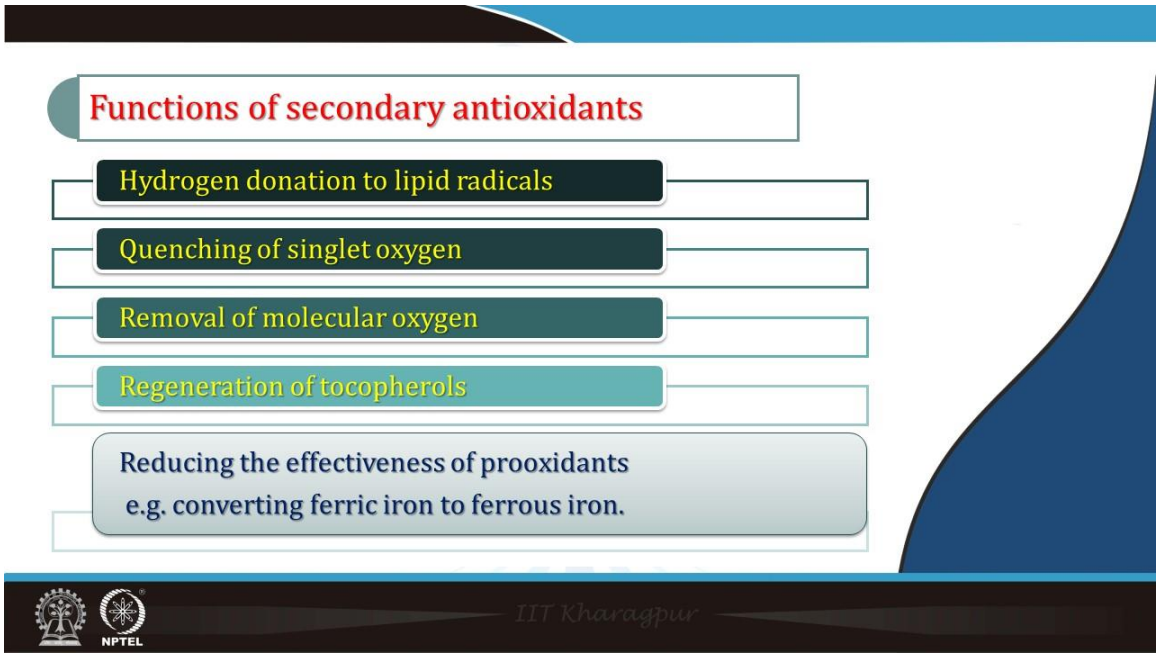


So, let us see the. Let us a bit, that is. The primary oxidants. They are basically hydrogen donating antioxidants. They can donate hydrogen atom or electrons to free radical species as you can see here. That is, this antioxidant is, you can transfer the electron to the free radical, and then accordingly the this free radical becomes a non-radical product, ok. So, the free radical that is the chain mechanism is terminated, that is, how these primary antioxidants work.

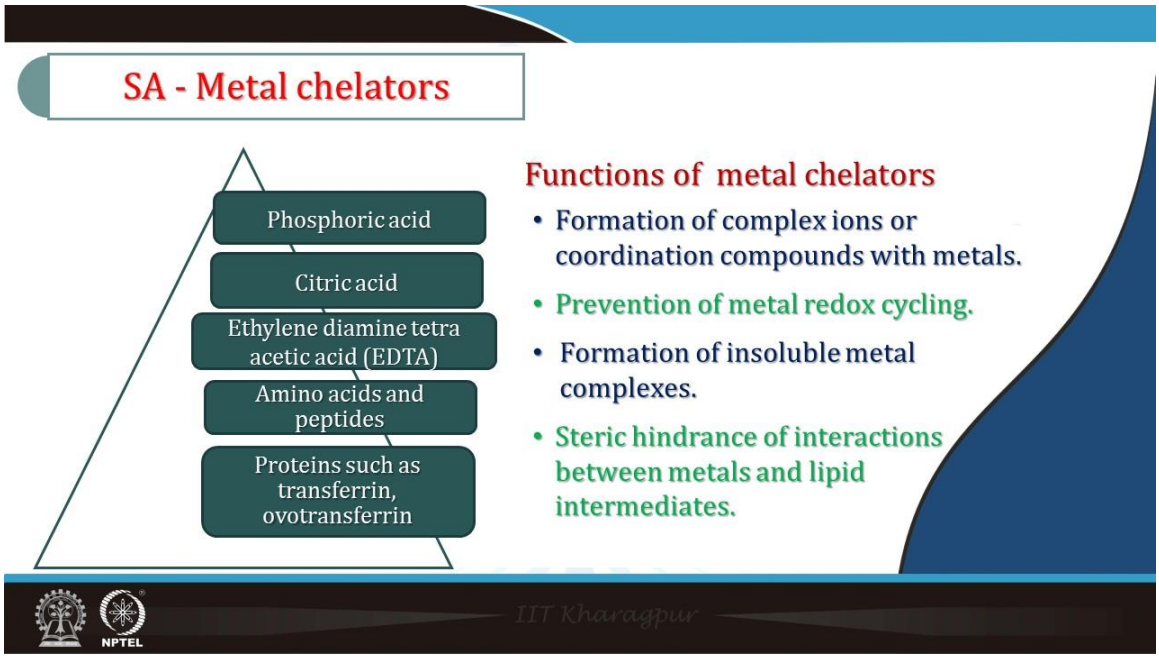
That is antioxidant free radical does not initiate another free radical due to stabilization of radical by delocalization. The primary antioxidants which are mostly the synthetic in nature, but they are extensively used in the oil processing, in the oil, refined oil whatever you buy from the market etcetera. You will find one or the other or even more than one antioxidants being used there, because these antioxidants mostly, we will discuss little later also, they are mostly, they are the synergistic, that is, if the two or more antioxidants are used together, their effectiveness is increased.



So, the butylated hydroxy anisole, butylated hydroxy toluene, propyl gallate and tertiary butyl hydro quinone; these are; their structural formulas are given here. And they are most commonly used synthetic antioxidants in edible oils and fats.



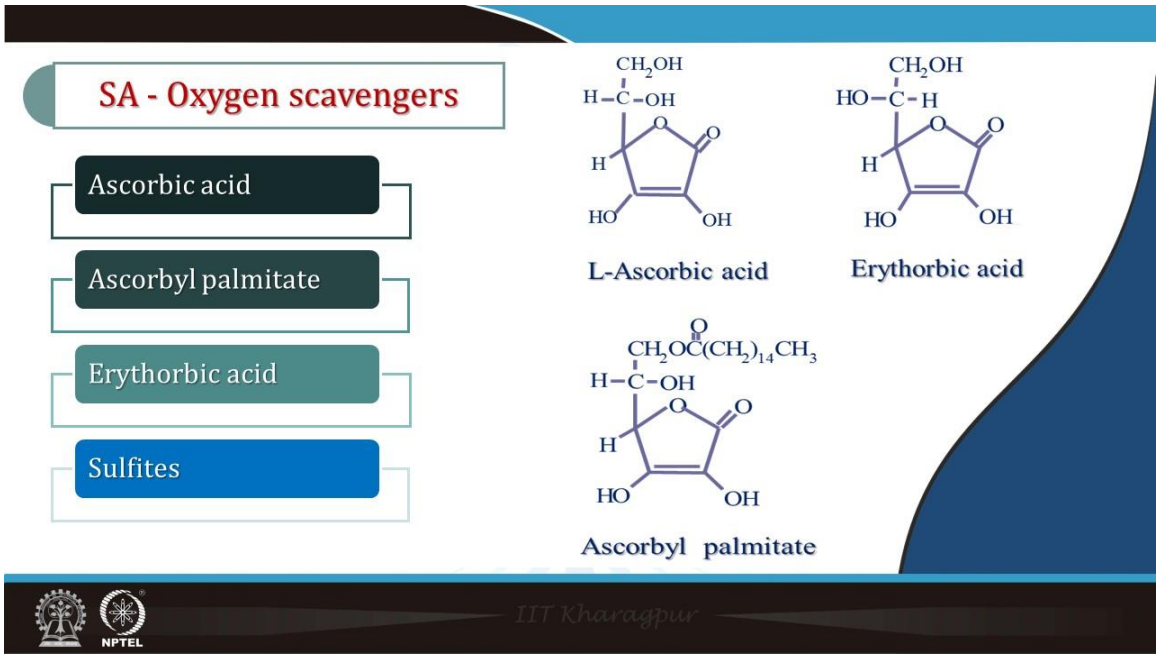
So, this secondary oxidants. How they work? That: they have the hydrogen donation to lipid radicals; they work by quenching of singlet oxygen; by removal of molecular oxygen and as well as regeneration of tocopherols, etcetera. That is the. Reducing the effectiveness of prooxidants like converting ferric ion to ferrous ion. So, this is how the secondary antioxidant function.



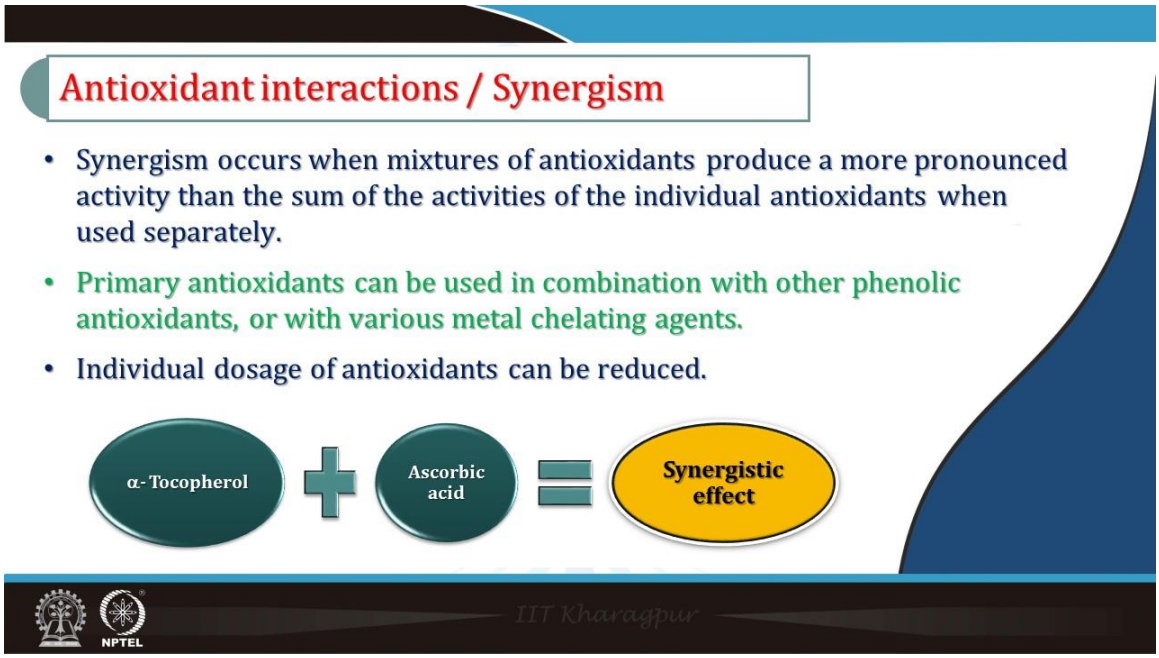
So, the metal chelators. We have seen they are the secondary oxidants. So, various metal chelators include phosphoric acid, citric acid, EDTA, amino acid, peptides, proteins such as transferrin, ovotransferrin, etcetera. These were; some of these we have discussed in the earlier slide as well.

So, how these metal chelators functions? That they form complex ion or coordination compounds with the metals, because, you see, in the most of the oils there might be some, either the chlorophyll linked etcetera to magnesium metal ion. There may be some during, the, when the oil is being treated during refining process or in the extraction. So, some metals may come into the oil from the machinery used, etcetera.

So, these metal ions, they, if they are allowed to be there they will obviously, oxidize. They will catalyze the autoxidation process. So, these metal chelators are used. What they do, that, they bind or chelate the these metal ions and then therefore, they block them or they prevent them from doing their, that prooxidant capacity activity, that is the from that working them as a activator of the oxidation process at catalyst of the oxidation process. So, they prevent metal redox cycling, formation of insoluble metal complexes like the steric hindrance of interactions between metals and lipids intermediate, that is, these process etcetera are there. So, they basically bind the metal ions and therefore.



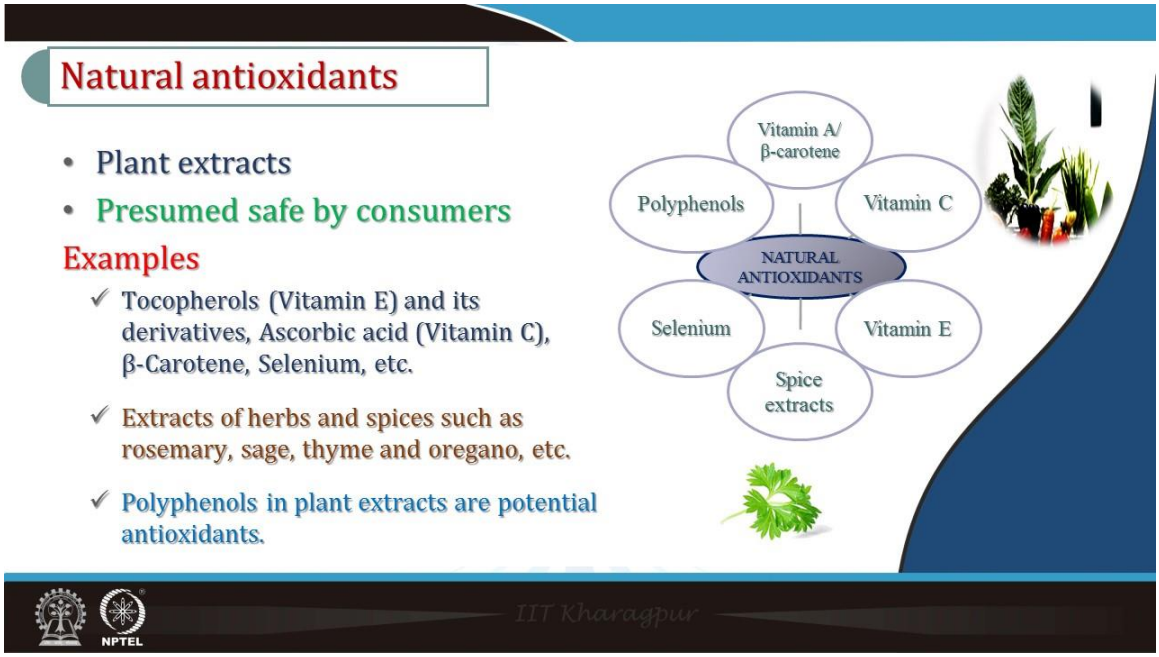
Then other group with the oxygen scavengers, that is, the secondary antioxidants like ascorbic acid, ascorbyl palmitate, erythorbic acid, sulfites, etcetera. Their structure is here given. So, they basically, they scavenge the oxygen, that is they, say, they remove the oxygen molecule, alright, from the environment, that is, then, they stop the reactions oxidation process, ok.



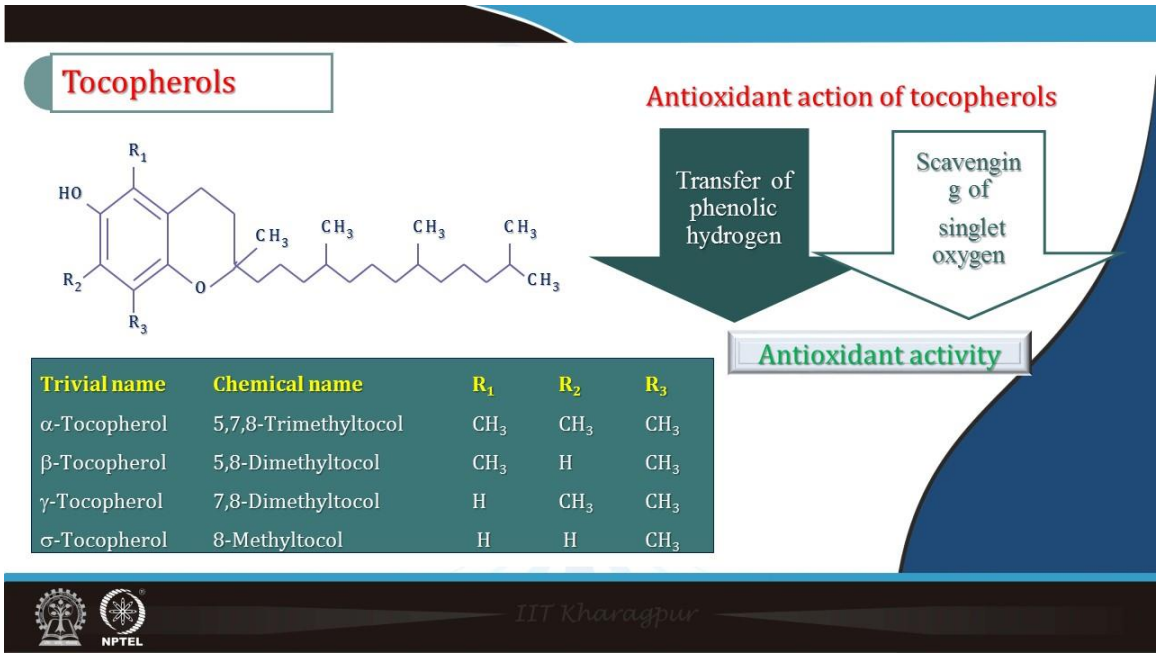
So, this antioxidant interactions or synergism. As I told you that is the various different antioxidants synergistic to each other, that is, synergism occurs when mixture of antioxidants produce a more pronounced activity than the sum of the activities of the individual antioxidant when used separately, ok. Like for example, that is if you are using antioxidant A and antioxidant B, separately. A has the activity X and B has the activity Y. Then if you use A and B together you will get another antioxidant activity Z which will be higher than A and B. That is the, what called synergism.

So, primary antioxidants can be used in combination with phenolic antioxidant or with various metal chelators, etcetera. That is, if you use primary antioxidant and secondary antioxidants together, you get a better synergistic effect, like alpha tocopherol and ascorbic acid. Alpha tocopherol is a primary antioxidant; ascorbic acid is a secondary antioxidant. So, if you are; if these two has a synergistic effect and when they are used in

together they have the give the better result in the system ok. Then now these were some of the primary and secondary antioxidant, but they were mostly of the synthetic origin.

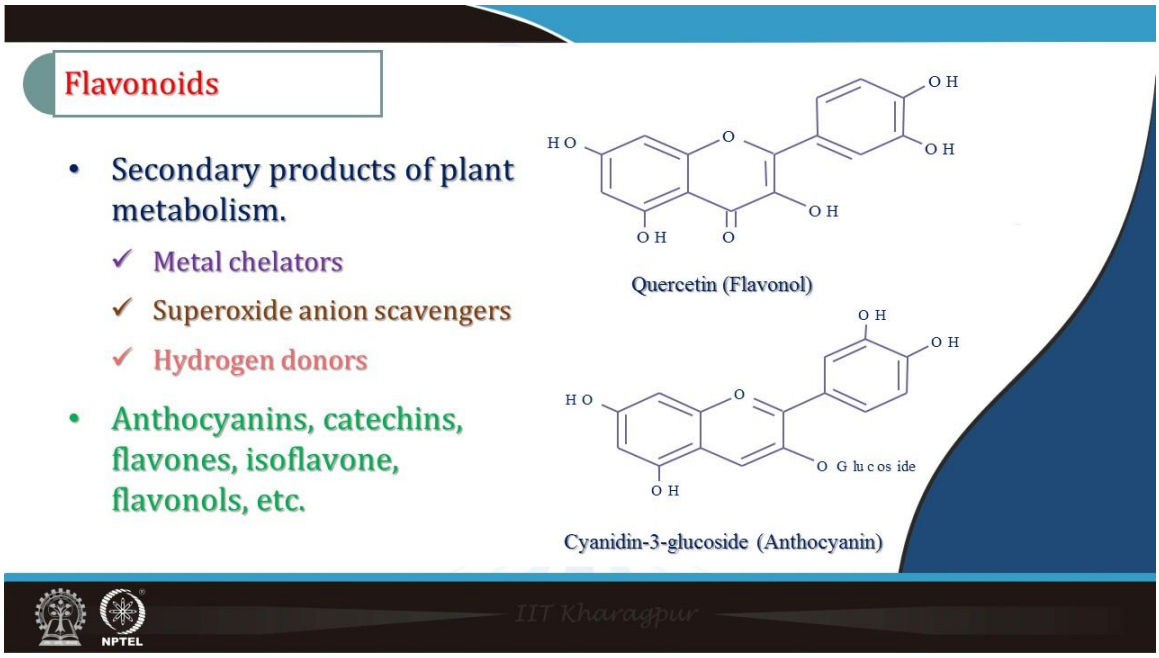


But now let us discuss about natural antioxidants because they are the various compounds like polyphenols, vitamin A, beta carotene, vitamin C, vitamin E, even selenium, spices extracts, etcetera. They are present normally naturally in the various plant and they can be extracted from these plant materials and then they can be purified or used as a extract, etcetera. And then. And these are presumed to be safe by the consumer because in the recent past particularly there were lot of literatures, and where the some undesirable effect on the health of the consumers, when excessive amount of the synthetic antioxidants, etcetera were used, they are reported. So, the, their consumers are now well concerned that they want more and more use of that is a natural antioxidant rather than going for synthetic antioxidants.



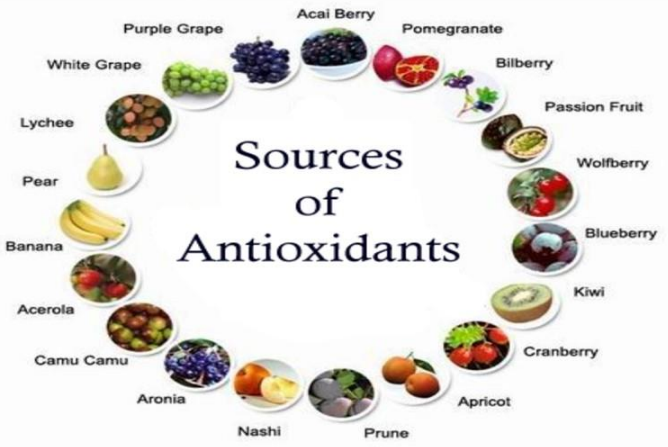
So, the you know the tocopherol, vitamin E and its derivative these are naturally present even that is in the pulses, oil seeds etcetera ascorbic acid is present in the fruit juices, beta carotene, selenium and so on. Extracts of herbs and spices like rosemary, sage, thyme and oregano they contain various phenolic compounds and such other compounds which have very good antioxidant activity. So, the tocopherols in plant extracts are potential antioxidants ok. So, the tocopherols. This is the structure of the tocopherols and here, that is a, it is a, you see, R_1, R_2 and R_3. So, depending upon whether this R_1 and R_2 and R_3, they are other methyl group or formyl group or combination of that, it becomes alpha tocopherol, beta tocopherol, gamma tocopherol and delta tocopherol.

And all this, R_1, R_2, R_3 are, as there is methyl group, it will become alpha tocopherol and accordingly it is given here. Then all this, there is the two are the methyl and one is the formyl, it becomes your beta tocopherol and so on. So, these, this and tocopherols. They are antioxidant action, that is, they are transfer phenolic hydrogen as well as scavenge a singlet oxidation as a singlet oxygen. So, by these both, they function their antioxidant capacity is by antioxidant function mechanism function is by transfer of phenolic hydrogen or scavenging of singlet oxygen.



Flavonoids. They are mostly the secondary products of plant metabolism. They acts as both metal chelators, superoxide anion scavengers and hydrogen donors. The anthocyanins like catechin, flavanones, isoflavones, flavanols, etcetera are examples of these major flavanoids which has good antioxidant capacity. And they are present in many spices and herbs etcetera and they can be extracted from there. And they can be used like quercetin, flavanol or cyanidine 3-glucoside (anthocyanin). These are. Their structural formula is given here. So, they can be.

Fruits and vegetables are excellent sources of natural antioxidants

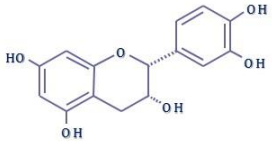


- Fresh fruits such as apple, pears, grapes, citrus fruits, etc.
- Vegetables like potato, tomato, spinach, carrot, etc.
- Contain various polyphenolic compounds.
- Wine is a good source of anthocyanins.

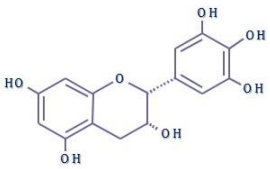
Source: www.biosciencenotes.com

Then fruits and vegetables are the excellent sources of natural antioxidants like fresh fruits such as apples, pears, grapes, citrus fruits, etcetera or vegetables like potato, tomato, spinach, carrot. Certain. They. All these, they, contain various phenolic compounds and wine is basically a very good source of anthocyanins. So, obviously, it is said that is the wine consumption ofcourse, there is in control amount it goods it has a very good antioxidant capacity ok.

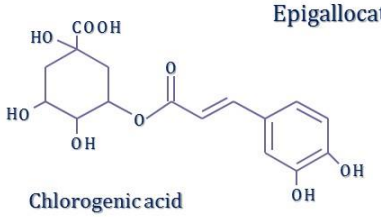
Antioxidants in tea and coffee



Epicatechin



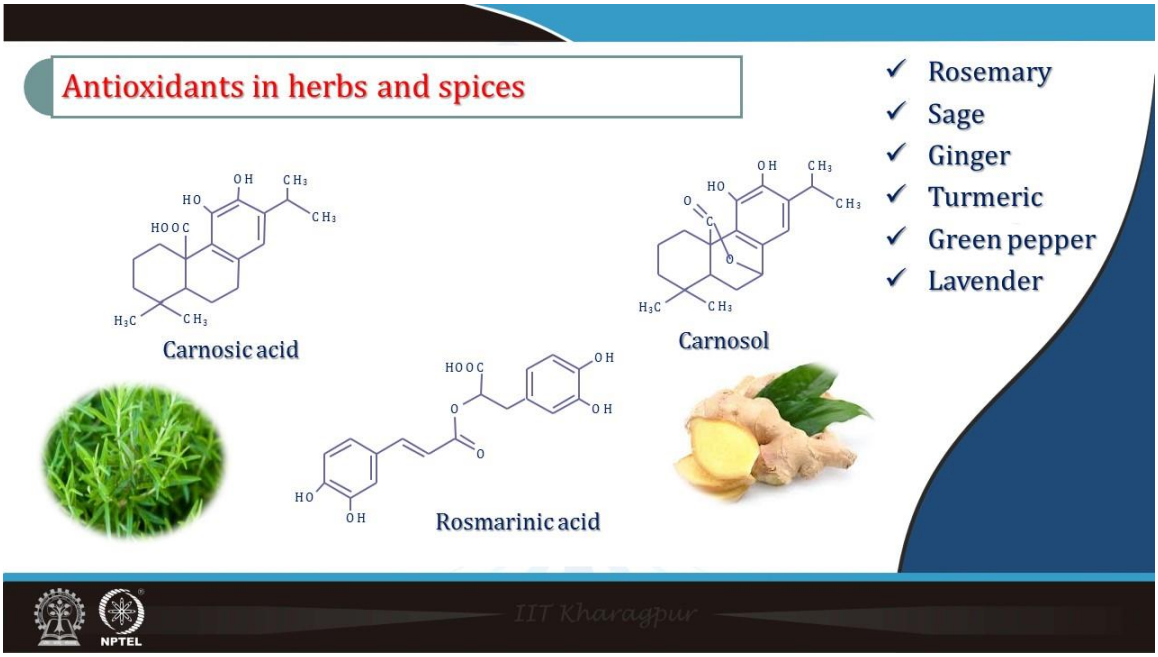
Epigallocatechin



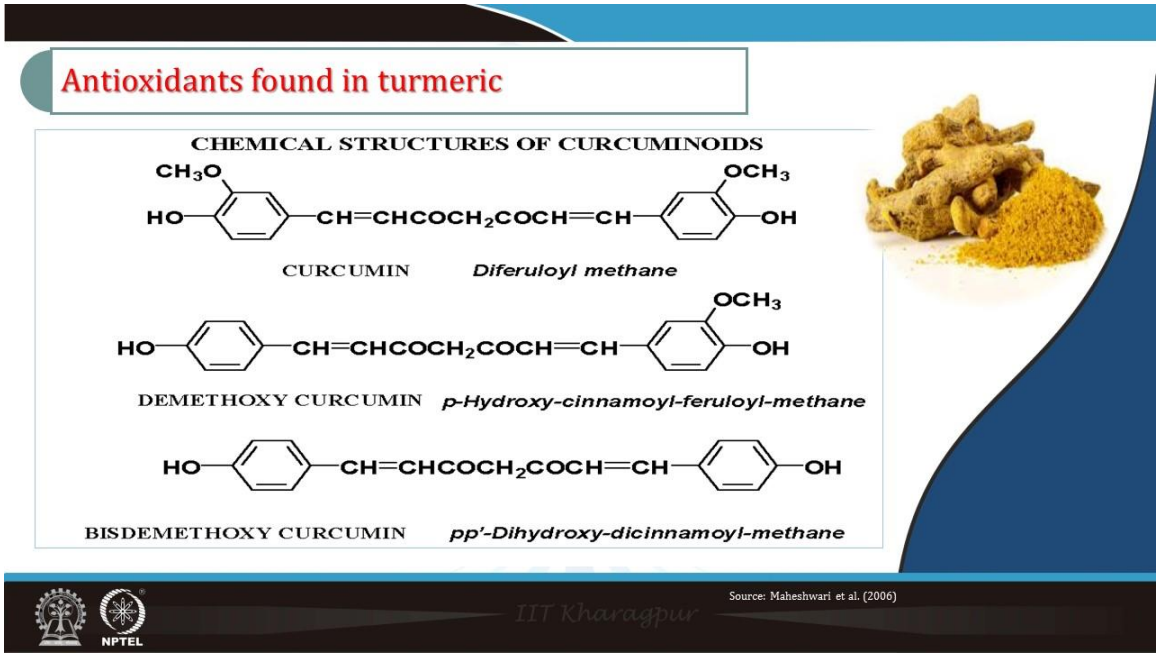
Chlorogenic acid

Then in the tea and coffee also this epicatechin, epigallocatechin, chlorogenic acid, etcetera are the primary source of the antioxidant capacity. And you see, what happens in the. They are present: epicatechin or epigallocatechin, etcetera mostly they are present in the green leaf. When these green leaves are converted into or processed into black tea then epicatechin or epigallocatechin are converted into that theaflavin as well as polymeric theorubigin. And their antioxidant capacity is little reduced, although they have other health promoting capacity activity. So, but in the green tea when we process green tea these epicatechin or epigallocatechin, they remain intact because in the black tea production by enzymatic action they are oxidized into thearubigin and theaflavin and theorubigin, ok.

And that is why, it is said that as green tea they have they are good as an antioxidant they have more health value than that of the black tea because this is mainly because of the these epicatechin, epigallocatechin these are the chlorogenic acid which are the phenolic substances and the polyphenols etcetera which are present intact. And of course, there one has to see that experiment and these juices that is if you take that is fresh tea leaf juice, that is, this can be used in the oil as an to prevent the autoxidation process. However, it is used has to be optimized, etcetera, that is, so that the doesn't interfere with the oil color, flavors and so on.

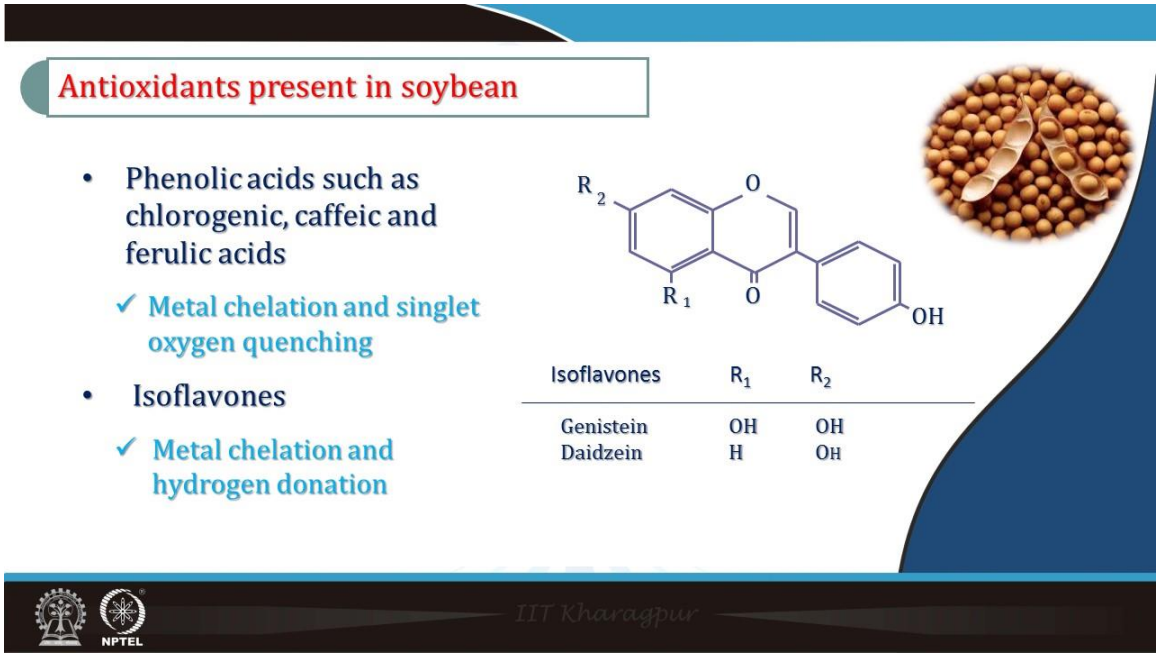


Then antioxidants in herbs and spices like rosemary, sage, ginger, turmeric, green pepper, lemon, etcetera they are the hub of various compounds like carnosic acid, carnosol, rosemarinic acid and so on. In fact, they have good antioxidant capacity we at IIT Kharagpur in our laboratory conducted a project where that rosemary and sage extracts were used in the oil alright in various proportions and we saw that our result indicated that use of these extracts rosemary and sage extracts which extended the shelf life of the oil not only the that is room stability, but also it improved thermal stability of the oils. So, this that is extracts of herbs and spices they have a great potential for use in oil.



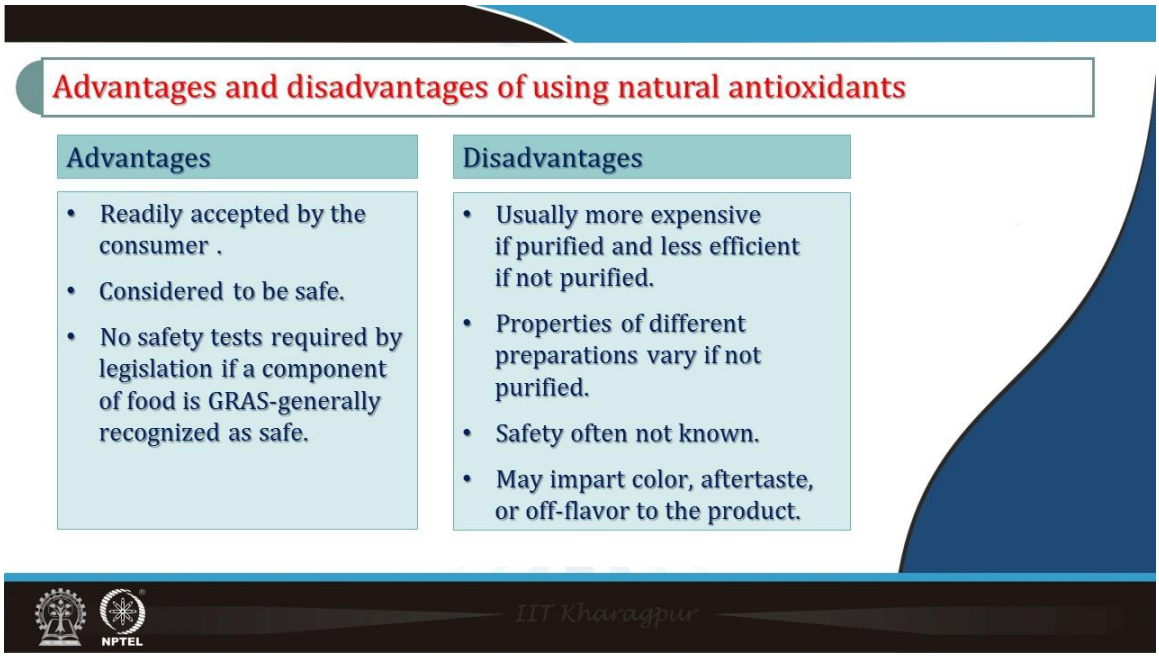
And even the turmeric that is this turmeric the antioxidants which are found are there is a curcumin that is the or dimethoxy curcumin or dimethoxy curcumin etcetera that is this curcumin that is these three are the major compounds curcumin, dimethoxy curcumin and bisdimethoxy curcumin. Their structure is given here and they again are very good source of antioxidant capacity. This also in our lab we used to increase the shelf life of the oil as well as that is because they can be used not only as a natural colour, but the natural antioxidant and their colour of these compounds also goes very well with the colours of the oil. Then the frying stability, frying quality of the products where this in the oil where these antioxidants that is the healthy they extracted there is these

compounds that is the extracts of turmeric or extracts of rosemary the things when we are used the both the oil stability as well as room stability as well as thermal stability, oxidative stability of the oil improved significantly ok.



Then this phenolic acids such as chlorogenic acid, caific acid, ferulic acid etcetera they are the antioxidant major antioxidant present in soybeans and they have metal chelating capacity that is they chelate the metals and they also scavenge or quench singlet oxygen ok.

So, these are the isoflavones structure is given here that is the it is a rings and R_1 and R_2 are here the so, flavones rings, ok. And there are two that is genstein and daidzein that is when R_1 is OH and R_2 both are OH it is a genstein and when R_1 is H and the R_2 is OH it becomes daidzein and accordingly that is this even antioxidant capacity of these two compound it is varies because of this presence of compound.



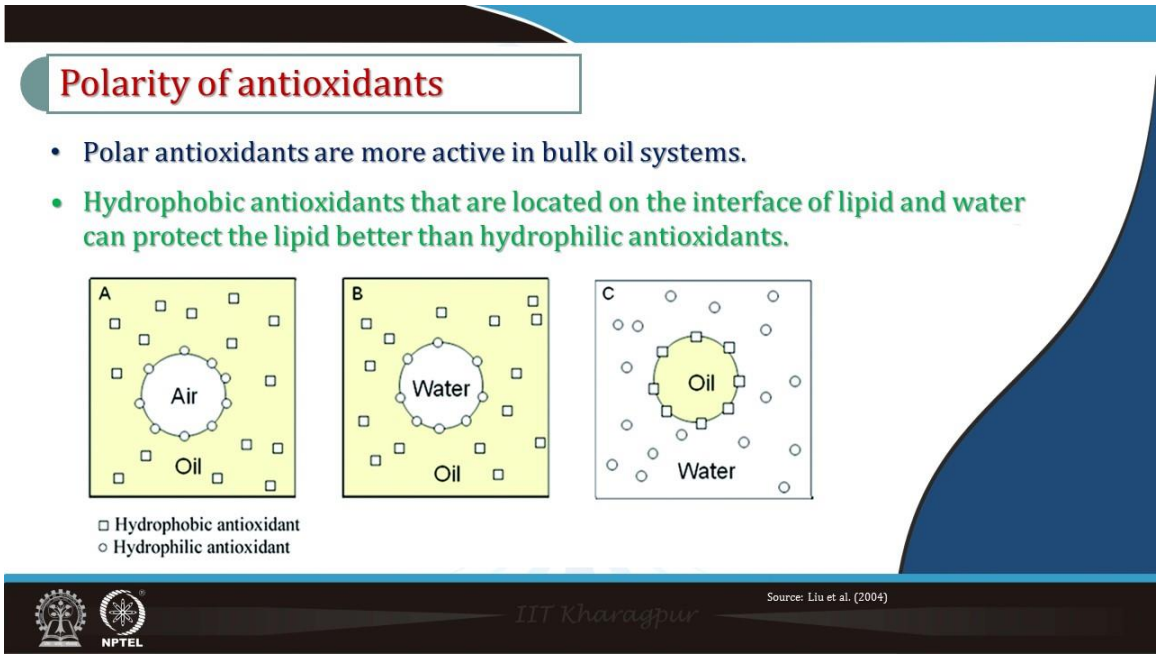
Now that there are both there is one natural antioxidant you use although there is a consumer trend nowadays trend in the market to use more and more natural antioxidant, but it has to be used in caution with caution because they are both advantages and disadvantages of using natural antioxidants. The advantages obviously, are readily accepted by the consumers they are considered to be safe and there are no safety test required by regulation if a component of food is a grass that is generally recognized as safe that is the chemical. So, it can be used you can simply that is if you are using a plant extract, a herb extract etcetera in the oil you can.

So, it does not invite any legal complications. However, there is may be that using that natural antioxidants may be usually more expensive ok if purified form and less efficient if it is not purified. So, that is a that is one problem with the or one challenge with the using natural antioxidant because if you take the herb extract. So, along with the compound that is in the extract the antioxidant concentration may be very very very less and they are not that effective. Then if you take the natural compound extract the natural compound from these that is extracts ok, but if you go for extracts and purification process it becomes the process becomes very expensive ok.

And also there is another problem that is if you go for purification you will like the particular rosemarinic acid etcetera that is if you this use in the rosemary extract, but

when you take it out purifies then it will can it is treated as a chemical and in that case one has to go and take the approval of the competent that is the legal agency or regulating agency in the country. So, also properties of different preparation vary if not purified ok. If not purified so, may be that you have to that is what is the concentration and what is the purification level that is unpurified form the concentration may vary at the compound may vary and the concentration may vary accordingly the its effectiveness may vary ok. And also and the unpurified form when you are you having the herbs extract or spice extract it may have some it is not variable because there are several compounds and might be some compound which are which might have adverse effect on the health that is so, safety always is not very much known about these natural antioxidant extracts etcetera ok. And also there is the if you want to use these natural antioxidants unpurified form particularly then the concentration of the extract or proportion of the extract quantity of the extract which is which might be required to be used in the oil to get the desired effect all right may impart color it may include some after taste or off flavor to the product.

So, these are some of the disadvantages of using natural antioxidant, but all these thing can be taken care there is a trend toward use of natural antioxidant.

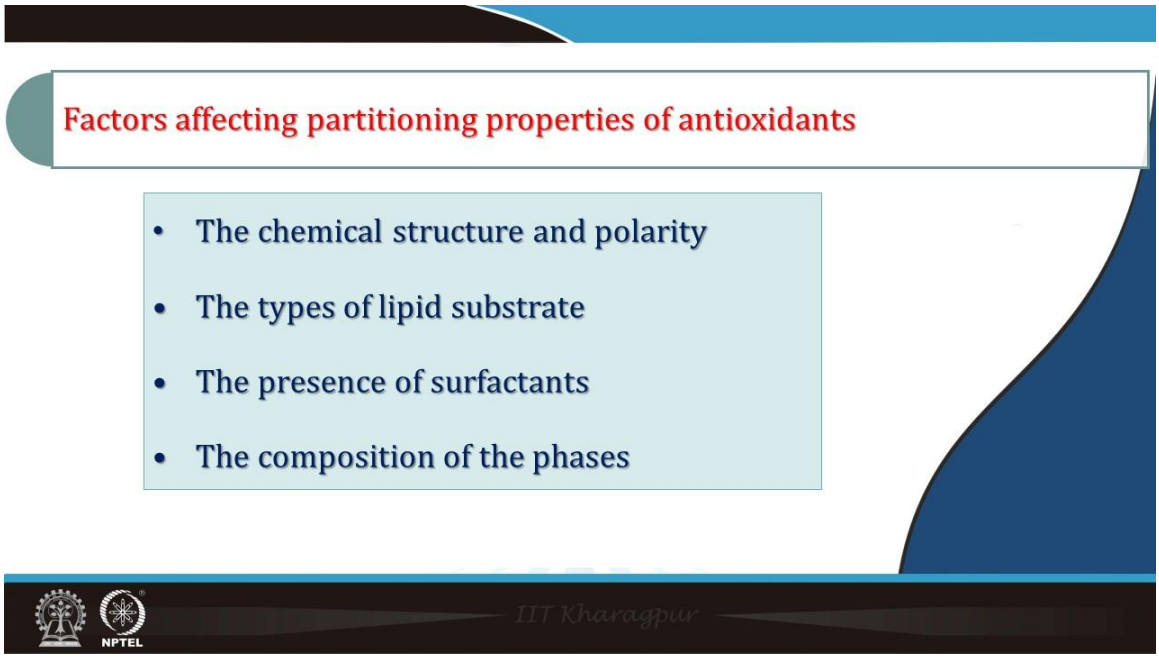


Then another thing is the polarity of antioxidant is very important that is the polar antioxidants are more active in bulk oil system ok. That is hydrophobic antioxidants that

are located in the interface of the lipid and water that is you can see that lipid and there is water molecule and the lipid oil ok. Then they can protect the lipid better than the hydrophilic antioxidants which are basically present in the water etcetera that is there you see that is the these are the hydrophilic antioxidants which are in the water ok. Then hydrophobic antioxidant these are which they are in the oil present in the.

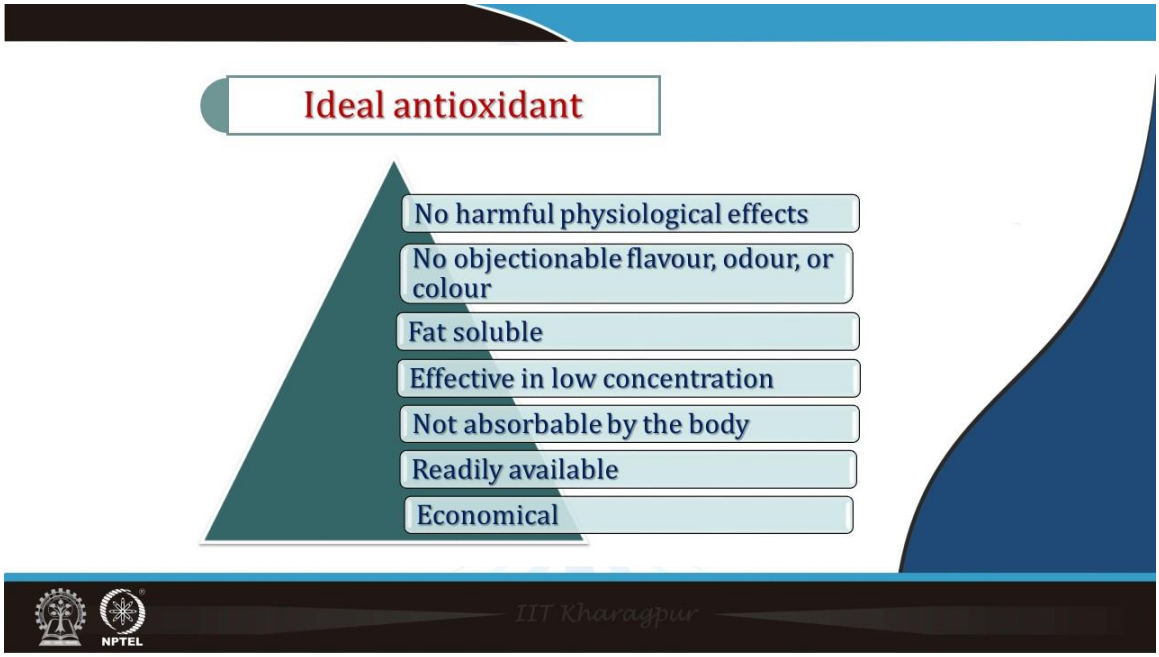
So, this here is the oil water in oil emulsion basically in the this B. So, this it is surrounded by also obviously, antioxidant is present in the oil water interface hydrophobic. So, they can protect the oil better that is ok. And there can be one other that is air where this oil and this contains both. So, air in this is the hydrophilic sorry hydrophilic antioxidants are present in the air.

So, this figure shows that yes this hydrophobic antioxidants are better more effective.



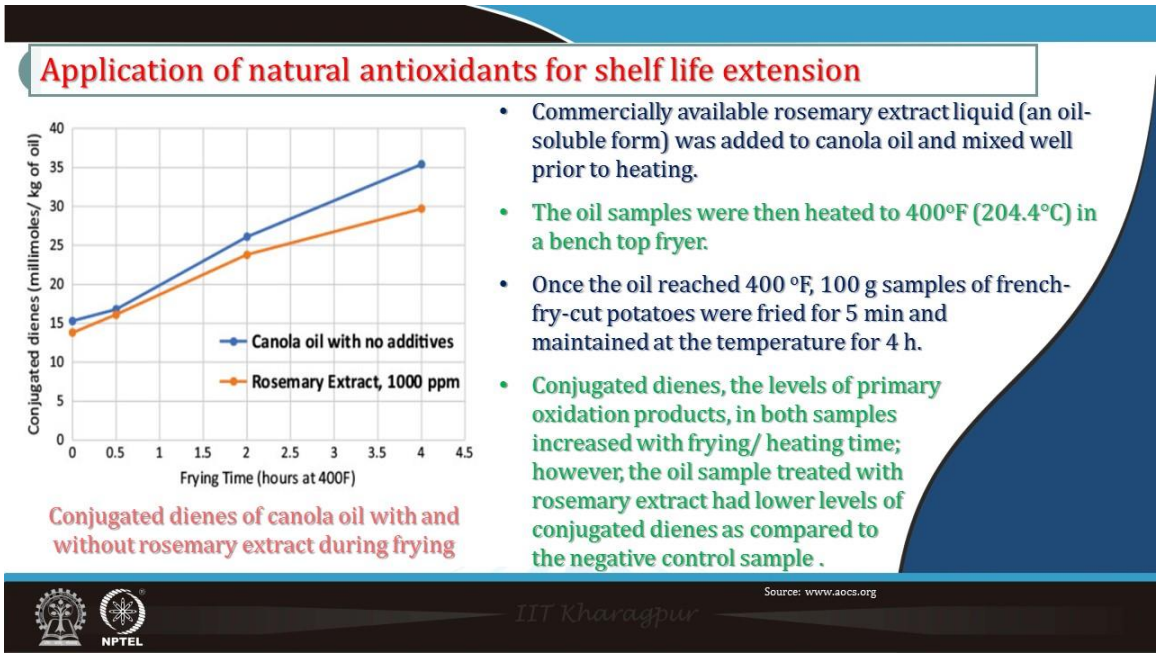
Common factors that affect the partitioning properties of antioxidants obviously, is the most important factor chemical structure of the compound that is polarity. Then the also the type of the lipid substrate what is it is a this type of fatty acids which are present there is that they are saturated fatty acids, unsaturated fatty acids, more polyunsaturated fatty acids all these will influence that the antioxidant properties use

doses required of the antioxidant type of antioxidant what is even the presence of certain surfactants are the composition of the various phases while water lubricant all these phases they will influence the partitioning properties of antioxidants ok.



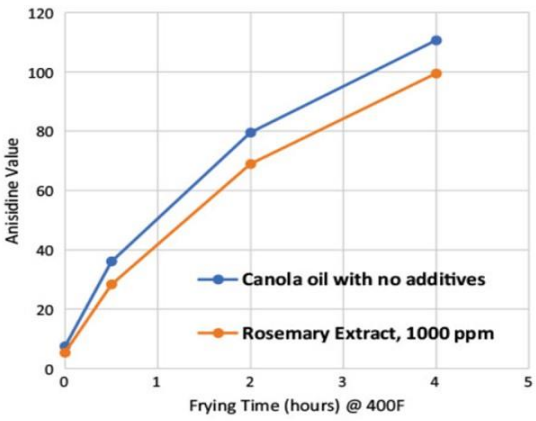
So, the ideal antioxidants we have said what should be if you are in oil business. So, and you want to use an ideal and I want to use antioxidant and you have to use if you want to make it more stable room temperature stable as a thermostable oxidation stable then antioxidants are required to be used.

So, obviously, for selecting ideal antioxidant one should give this a consideration like it should have no harmful physiological effect, there should be no objectionable flavor, odour, or colour etcetera when they are added in the required proportion the antioxidants should be fat soluble. So, that it easily mixes with the oil it should be effective at low concentration particularly its antioxidant potential antioxidant capacity should be at low concentration it should not be absorbable by the body readily it should be readily available and obviously, it should be economical right.



Then these are the application of natural antioxidant for shelf life extension some case studies also like the conjugated dyes of canola oil with and without rosemary extract is as we seen here that is the here the yellow lines are showing that is where the rosemary extract is having 100 ppm and the blue lines there is no rosemary extract. So, you can see conjugated dry in formation is reduced in the oil which has rosemary extract ok. This commercially available rosemary extract like liquid and oil soluble form in this experiment was added to canola oil and mixed well ok.

And these oil samples were heated to about 204 degree Celsius in a bench style bench top fryers and once the oil reached that 100 gram sample of French fry cut potatoes were fried for 5 minute and maintained at that temperature for 4 hours ok. And then it these samples were analyzed for conjugated dyes formation and they are the labels in the you see that these are the primary oxidation products and they were found more in the oil which has no added rosemary extract ok. So, this we have taken from the other refers, but similar work we also did in our laboratory and the it gives similar effect ok. Here it is the another case is to the like canola oil ok, which having no additives or it is having additive rosemary extract 1000 ppm ok.



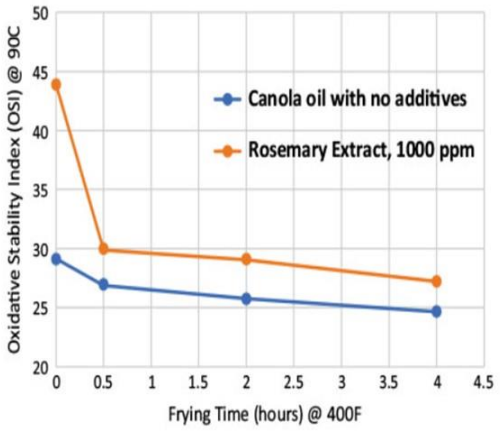
Anisidine values of canola oil with and without rosemary extract during frying

- Anisidine values, a measurement of the secondary oxidation products of lipids, indicated that the rosemary extract-supplemented oil sample had lower levels of anisidine values during frying/heating than the control oil sample
- This indicates that oxidative stability of the oil improved by adding rosemary extract.

Source: www.aocs.org

That is the and in this case an acid in value was measured it is a measurement of the secondary oxidation products of lipid and this indicated that the rosemary extract supplemented oil sample that had lower levels of an acid in value during frying or heating than the control sample.

This indicates that the oxidative stability of the oil was improved by adding rosemary extract which I told earlier as well ok.



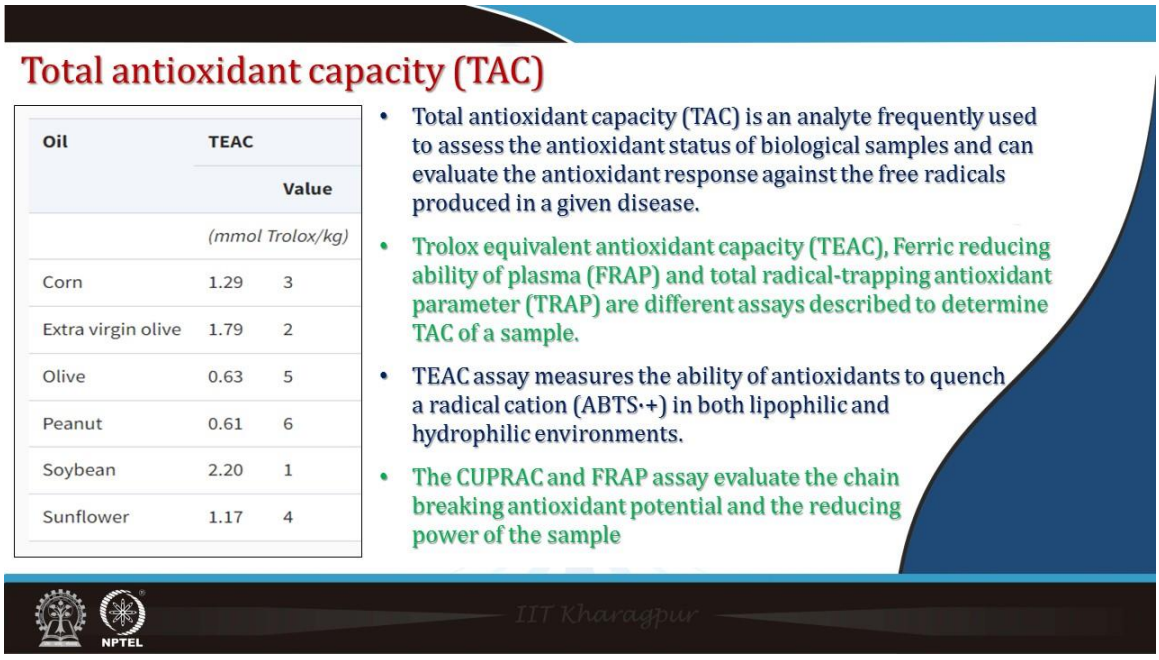
Oil Stability Index (OSI) of canola oil with and without rosemary extract during frying

- The oil stability index (OSI) of samples collected during various stages of frying were measured with the Oxidative Stability Instrument according to the AOCS Official Method Cd 12 b-92.
- The OSI values of oil samples decreased during frying and heating, with the rosemary extract treated oil sample remaining significantly higher than the control oil sample.
- These data suggest that rosemary extract treated oil sample had a longer "fry-life" than the control oil with no additives.

Source: www.aocs.org

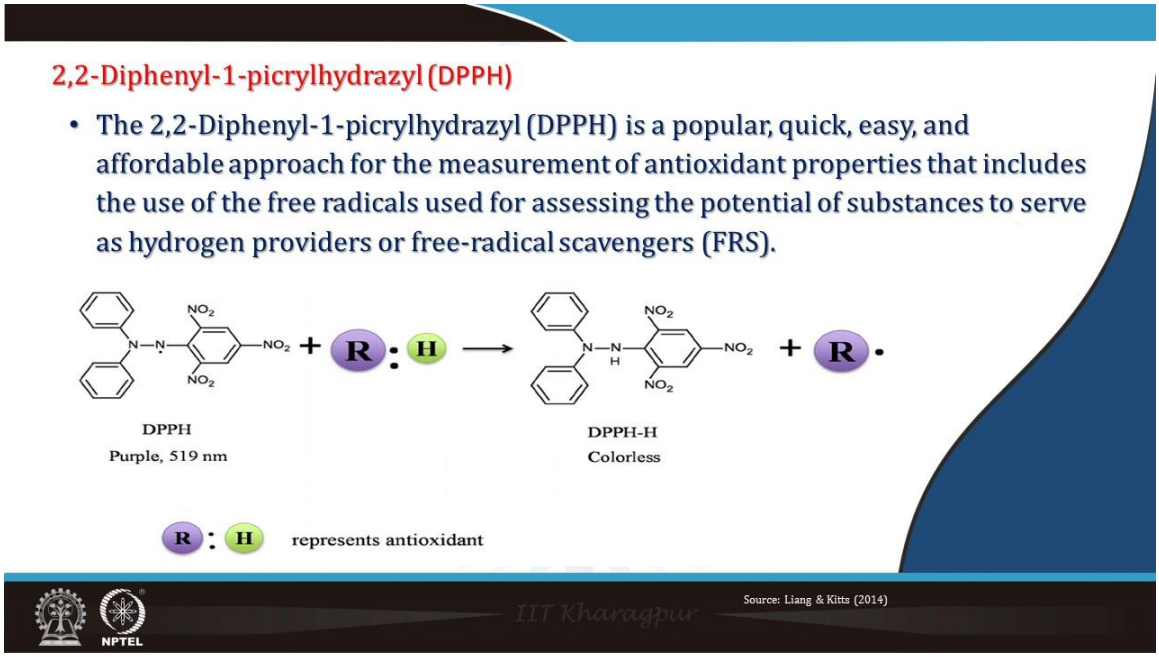
This is another case study where oil stability index was measured ok. And the samples which were collected during various stages of frying that is that using canola oil samples one was having no additive, other sample had rosemary extract 1000 ppm and then during various oxidation in the frying process the samples were taken and the samples were analyzed for oxidative stability using the instrument and give standard AOCS protocol. And the OSI values of oil samples decreased during frying and heating and particularly those which were having rosemary extract ok. And this data suggest that rosemary extract treated oil sample had a longer fry life that it has a better heat thermal stability than the control oil with no additive.

So, these three case studies this is a canola oil and this is a oil we can say that yes the plant extracts are rosemary sage and such other extracts they which have natural antioxidants and they have better potential for use in the oil. In fact, when you are frying like potato chips or such other fried products when it is being fried in the oil then in fact, if you assume turmeric ok. So, the yellow color of this turmeric it also improves the color it gives the very golden delicacy brown color of the fried product and which is many a times liked by the consumers golden product.

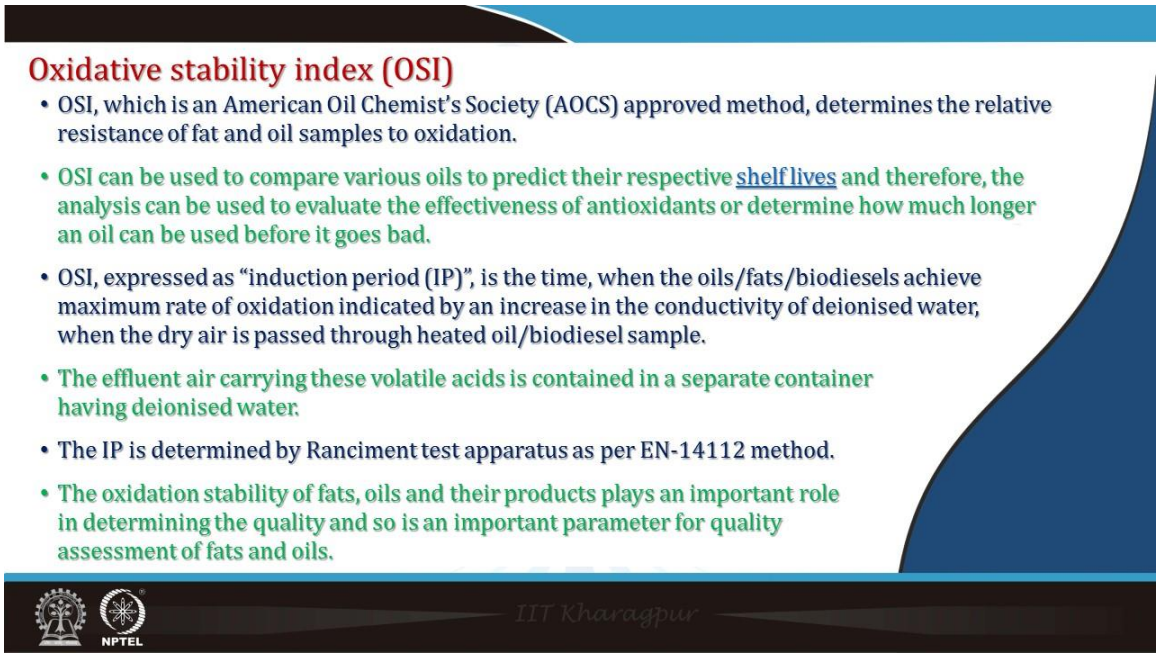


So, the total finally, the total antioxidant capacity of a compound of a product that it is an analyte frequently is used to access the antioxidant status of biological samples

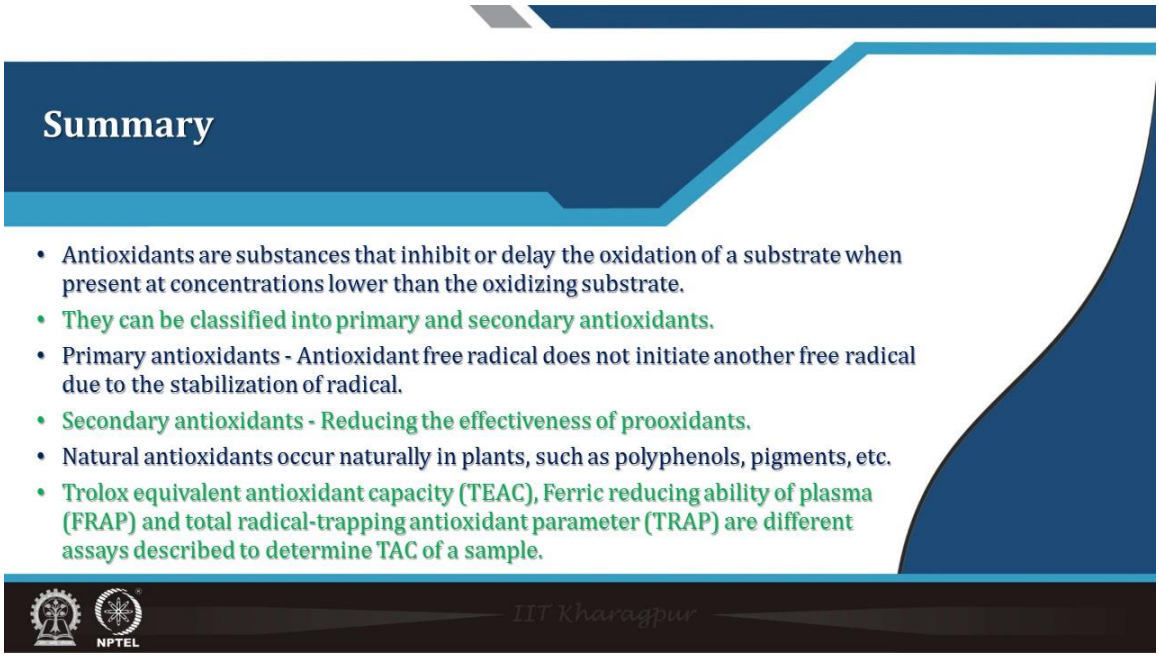
and can evaluate the antioxidant response against the free radicals produced in a given disease. So, torolox equivalent antioxidant capacity like TEAC or ferric reducing ability of plasma like FRAP and total radical trapping antioxidant parameter TRAP are different assays which are described to determine the total antioxidant capacity of a sample. TEAC assay measures the ability of antioxidant to quench a radical cation in both lipophilic and hydrophilic environments. The CUPRAC and FRAP assays evaluate the chain breaking antioxidant potential and the reducing power of the sample ok. Here some of the values of the TEAC and TRAP values are given in the some common oils ok.



Then two two diphenyl picryl dihydrogen that is a DPPH assay which is a it is a popular quick easy and affordable approach for the measurement of the antioxidant properties that includes the use of a free radical used for assessing the potential of substances to serve as a hydrogen providers or as a free radical scavengers FRS. So, you see DPPH that is R free radical RH ok, then it gives the NO_2 and gives a medical that is R free radical ok. And with this that is here we can say RH it represents this antioxidant this antioxidant. So, it gives R free radical and antioxidant come it protect the oil that is this is the antioxidant free radical is formed and it gives the DPPHH ok, which is a colorless compound. So, it gives the indicates that antioxidant potential of the oil.



Oxygen stability index which is a American oil chemist society approved method determines the relative resistance of fat and oil samples to oxidation. OSI can be used to compare various oils to predict their respective shelf lives and therefore the analysis can be used to evaluate the effectiveness of antioxidants or determine how much longer the oil can be used before it goes. But the oxygen stability index which is expressed as induction period commonly 'IP' is the time when the oils or fats or biodiesel etcetera achieve maximum rate of oxidation indicated by an increase in the conductivity of de-ionized water when the dry air is passed through heated oil or biodiesel sample. The effluent air carrying these volatile acid is contained in a separate container having de-ionized water and then IP is determined by the Ransiment apparatus Ransiment test apparatus as per the standard protocol ok. The oxidation stability of the fats, oils and their products plays an important role in determining the quality and so is an important parameter for quality assessment of the fats and oils.



So, finally, I would like to summarize this lecture by saying that these antioxidants are very very important substances as far as the oil milling industry is concerned. These antioxidants inhibit or delay the oxidation of a substrate when presented at a very lower concentration. They can be of two types primary and secondary antioxidants. Primary antioxidants that is these are the antioxidant free radical that does not initiate another free radical due to stabilization of the radical and secondary antioxidant they act by reducing the effectiveness of prooxidants. Total antioxidants occur naturally in the plant such as polyphenols, pigments etcetera and that is the TEAC, FRAP, TRAP etcetera are the efficient assays which are described to determine the total antioxidant capacity of a oil. However, these antioxidants they are used of course, this beneficial in the oil as far as the this oil stability is concerned preventing the oxidation, improving its thermal stability, oxygen stability etcetera, but it has that is both this antioxidants which are from synthetic origin. They are certain health concerns even the natural sources antioxidants they has to be used to they have to be used with proper care. So, that it is has to be used with proper care and proper control and monitoring in their use and effectiveness is very essential.

These are the references which are used in this class.

Thank you very much for your patience hearing. Thank you. Thank you.