Indian institute of Oilseeds Research, Hyderabad

Presentation on Professional Attachment Training

"Oil Quality Parameters and Screening on Antioxidants in Sesame Varieties"

(From 12-05-2015 to 11-08-2015)

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INDIAN INSTITUTE OF CHEMICAL TECHNOLOGY
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Part-1

Oil Quality Parameters

- ✓ Molecular Species
- ✓ Separation of lipid classes
- ✓ Mineral content
- ✓ AV, SV, USM, IV, PV

Introduction to oil quality

- ✓ Country production is 31 MT
- ✓ Area under production is 28 Mha
- ✓ Consumption is 16 kg/ annum/person (but 7-8 kg/annum/person)
- ✓ As consumption of oil increases import also increases
- ✓ Awareness for quality and safety also increased
- ✓ Oil has several acceptable quality parameters to accept
- ✓ Quality checks are fixed by ISI/BIS in India outside (AOCS, EU etc.

Molecular Species

Mix oil+Tris buffer+$CaCl_2$+bile salt

↓ Shake for 1 min at 40°C

Addition of lipase enzyme

↓ Shake for 3 min at 40°C

Add 1 ml of ethanol and 1.5 ml of 6N HCl

↓

Extract with EtOAc (2X5ml)

↓

Wash with water to neutralize and pass through sodium sulphate

↓

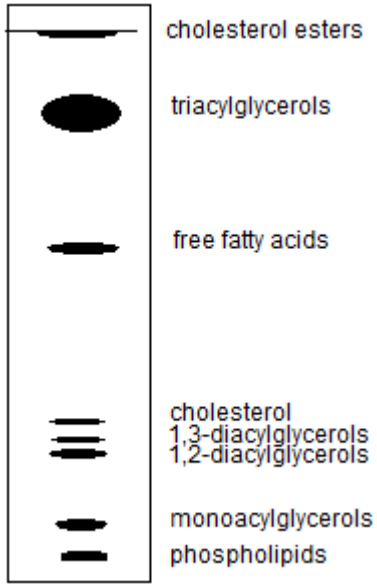
Concentrate the solvent and load for preparative TLC (H:EA:AcOH)

↓

Scratch MAG layer esterify to get FAME analyse by GC

ECN= TC-(2Xdouble bond)

Separation of Lipid classes of Oil

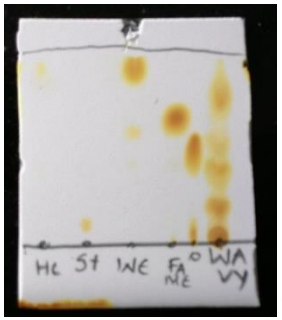


LIPIDS	Solvent used in column	TLC Solvent System
Neutral lipids	chloroform	Hexane : ethylacetate
Glyco lipids	acetone	Methanol : chlorofrom
Phospho lipids	methanol	Methanol : chlorofrom : water

Surface Lipids

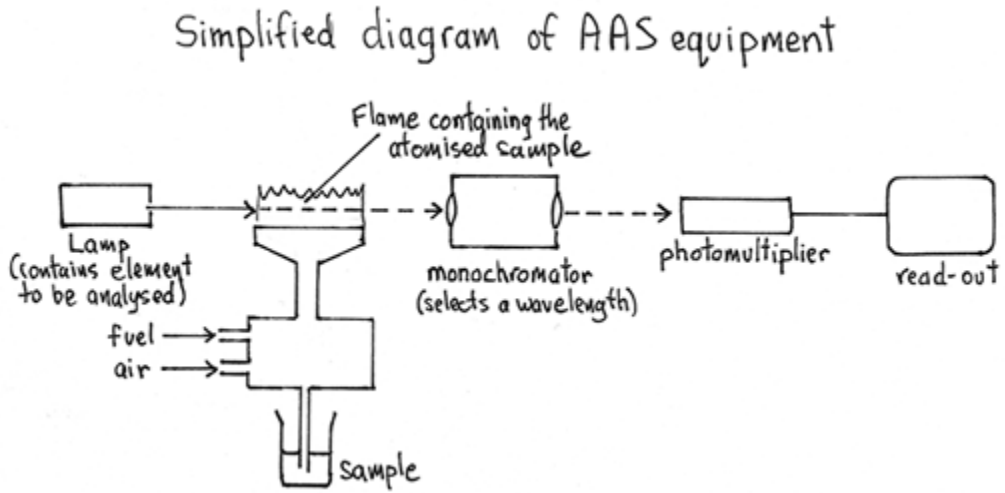
- ✓ Surface lipids are extracted by dipping plant material in chloroform for 30 sec
- ✓ Solvent is filtered and concentrated
- ✓ Obtained content is fractioned by column using different solvents
- ✓ Different fractions are obtained and checked by TLC in 100% to Toulene

Solvent used	Fraction obtained
Pentane	Hydrocarbons, wax esters
Hexane	Mostly sterols
1% EtOAc : Hx	FAME
2% EtOAc : Hx	TAG, DAG, MAG
5% EtOAc : Hx	Fatty alcohols, fatty acids
Chloroform : Methanol	Polar compounds



Mineral Content

- ✓ Digestion of oil/oil seeds using tri acid mixture
- ✓ Dilution with distill water
- ✓ Analysis by AAS
- ✓ Elements analysed are
- ✓ Ca, K, Mg, Na, Zn, Fe, Mn, Al, B, CR, Bi, CU, CO



Quality Parameters

- ✓ Acid value
- ✓ Free Fatty Acid
- ✓ Saponification Value
- ✓ Unsaponifiable matter
- ✓ Peroxide value
- ✓ P-Anisidine value
- ✓ Iodine value
- ✓ Hydroxyl value
- ✓ Moisture content

Using Instruments

- ✓ Colour by tinctometer
- ✓ FAME conversion by GC
- ✓ Tocopherols by HPLC
- ✓ Rancidity by rancimat
- ✓ Phosphorus estimation by spectroscopy
- ✓ Pesticide residue analysis by GC/HPLC

Techniques…..

- ✓ Refining of crude oil (degumming, neutralization, bleaching, dewaxing, deodorization)
- ✓ Biodeisel preparation
- ✓ Grease/lubricant preparation
- ✓ Wax preparation
- ✓ Resin preparation

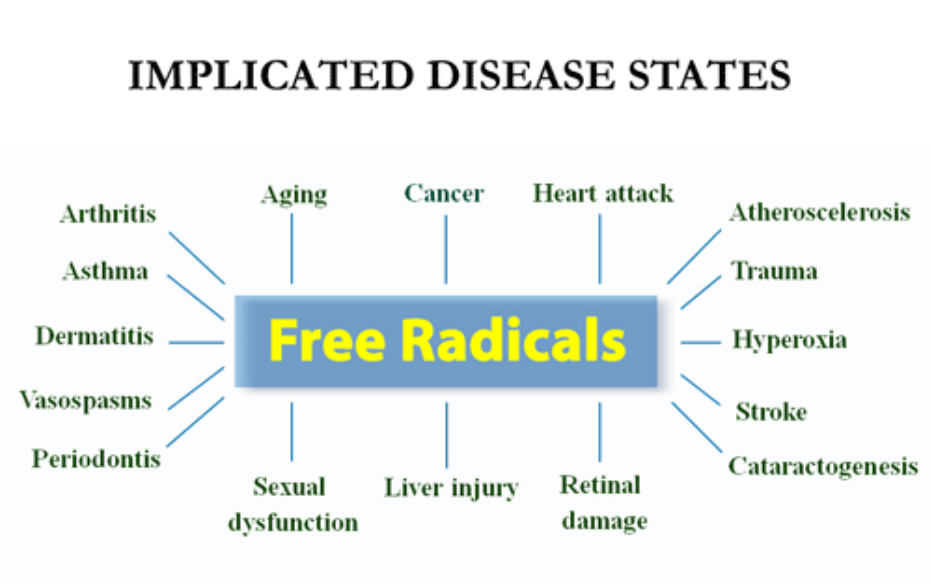
Part-2

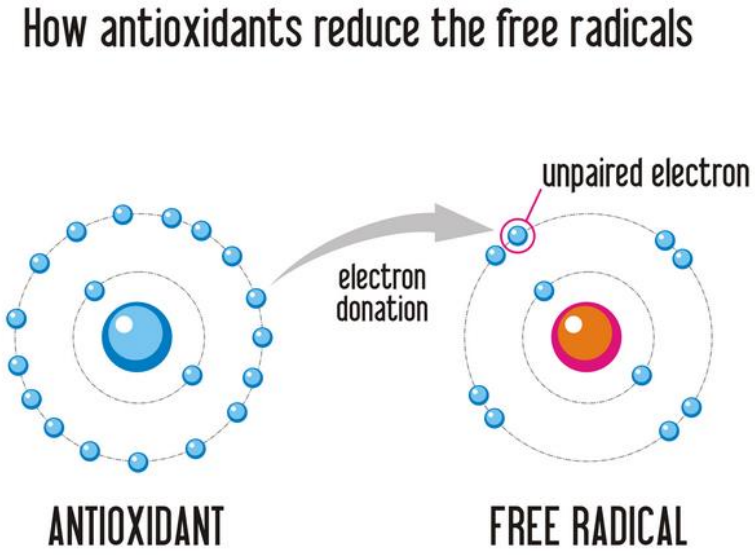
Antioxidants screening in Sesame

- ✓ Oil Extraction
- ✓ Fatty acid Profiling
- ✓ DPPH activity
- ✓ Phenolic content

Introduction

- ✓ Reactive oxygen species (ROS) react easily with free radicals to become radicals themselves





- ✓ WHO – 80% of the world populations rely primarily on traditional medicine as source for their primary health care

- ✓ Natural antioxidants can be phenolic compounds, carotenoids as well as ascorbic acid available in fruits and vegetables produced as secondary metabolites

Sesame

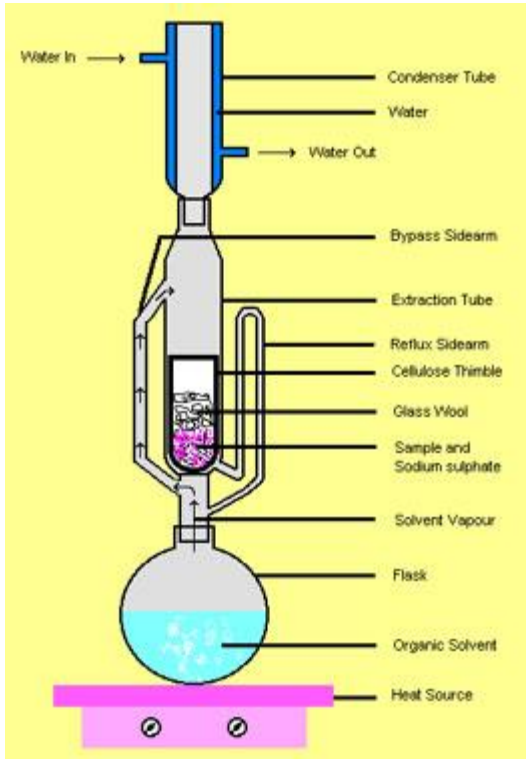
- ✓ India ranks first in world with 17.06 lakh ha area and 6.8 lakh tonnes production
- ✓ Average yield of sesame (402 kg/ha) in India
- ✓ *Sesamum indicum* L. belongs to Pedaliaceae family
- ✓ Sesame seeds contain many phytochemicals

Indian Varieties of Sesame

Variety	Place of growing	Year of release	Seed yield (kg/ha)	Days to maturity	Salient characters
Nirmala	Odisha	2003	800-900	80-85	White seed
G.Til-2	Gujarat	1994	750-800	88-92	White seed
G.Til-10	Gujarat	2002	750-800	88-92	Black seed
Swetha	Andhra Pradesh	1997	750-800	82-86	White seed
Hima	Andhra Pradesh	2006	800-850	80-85	Shiny white seed
DS-5	Karnataka	2012	600-700	90-95	White bold seed
Savitri	West Bengal	2008	1100-1400	84-88	Light brown seed
RT-351	Rajasthan	2010	700-800	80-85	White seed
JLT-408	Maharashtra	2010	700-800	80-85	White bold seed
RT-346	Rajasthan	2009	750-850	82-86	White seed

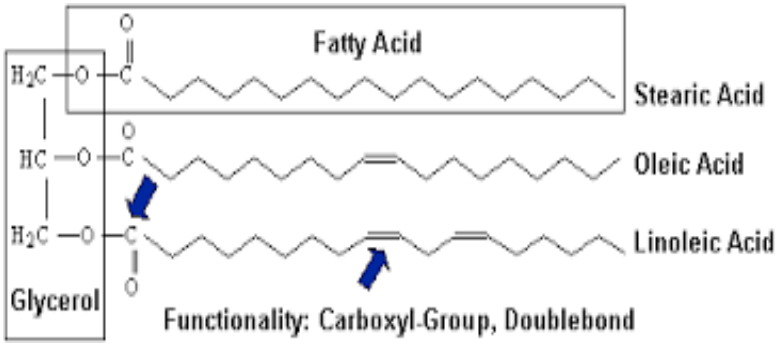
Oil Extraction by Soxhlet

- ✓ Hexane is used as solvent for extraction
- ✓ Oil content (%) = (Weight of oil obtained)/(Weight of sample taken) X100



Fatty Acid Profiling

- ✓ By FAME conversion and analysed by GC
  - ➢ Detector- flame ionization detector (FID)
  - ➢ Column-DB-225 capillary column
  - ➢ Temperature-160°C for 2 min; raised 220 °C at 6◦C /min
  - ➢ Inlet and detector -230 ◦C.



Calculated Oxidizability Value (Cox)

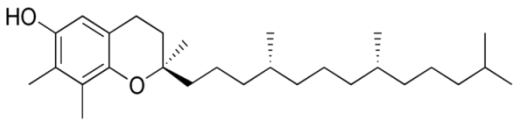
$$Cox = [1(18:1\%) + 10.3(18:2\%) + 21.6(18:3\%)]/100$$

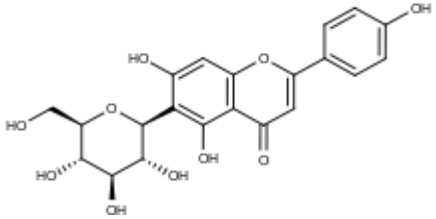
Extract Preparation from sesame seed

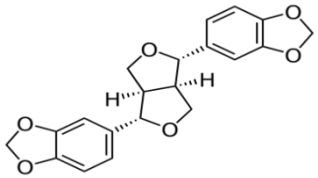
- ✓ Methanol + Sesame seed powder
- ✓ 8-10 hrs

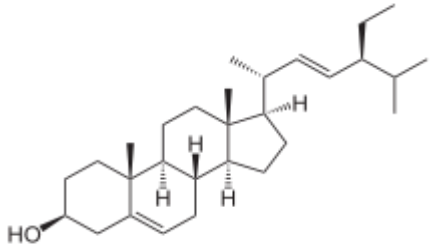
Identification of Antioxidants

- Detector- mass selective detector (MSD)
- Column-HP-1 capillary column
- Temperature-150°C for 2 min; raised 300 °C at 10◦C /min
- Inlet and detector -325 ◦C.
- MS library-Wiley library7n.1 and NSITO2.L





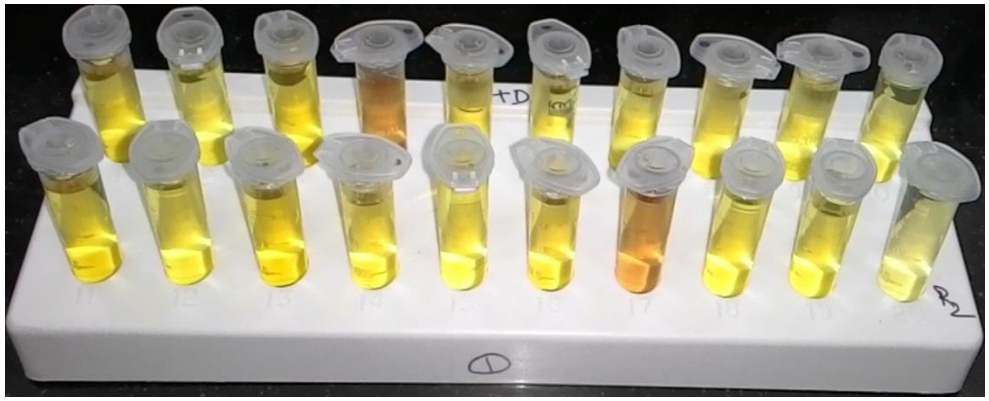




DPPH radical scavenging activity

% radical scavenging activity = 1-(Sample absorbance)/(Control absorbance) X100

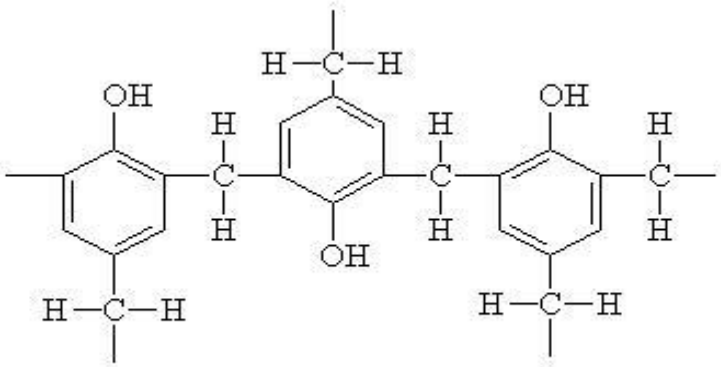
DPPH radical scavenging activity



Incubation period= 30 min
Wavelength= 515 nm
Reference solvent= methanol

Total Phenolic Content (TPC)

Incubation period= 15 min at 45°C
Wavelength= 765 nm
Reference solvent= Water



Phenolic

CONCLUSIONS

- ✓ Indian sesame varieties are rich in antioxidants
- ✓ High oil content was exhibited
- ✓ Studied parameters clearly states dependency on geographical conditions


long way ahead.....