Science Last Fortnight

Increasing Mango Shelf Life
Guar gum and essential oils

After harvesting, unripe mangoes rapidly turn yellow, due to increased ethylene production and rise in cellular respiration resulting in shorter post-harvest life. Much has been done to improve shelf life but these methods fail to preserve the taste of the mangoes.

Image: Ranjith-Chemmad, Wikimedia Commons

To overcome this problem, scientists from the University of Karachi, Pakistan, came out with a safer, consumer friendly solution: coating unripe green mangoes with a mixture of guar gum and essential oils. They carefully handpicked unripe green mangoes, based on uniformity of size and colour, ensuring that the fruit were free of physical deformities.

The team then extracted essential oils from seeds of fennel, nigella and coriander as well as from bay leaves, with ethanol and methanol, separately. And they mixed each essential oil with guar gum and glycerol in distilled water. These mixtures were homogenised by stirring and kept undisturbed for a few minutes.

They dipped unripe green mangoes in the coating solutions thrice at intervals of 10 seconds and allowed them to dry. After any excess coating solution dripped dry, these were stored in zip-lock bags in controlled temperature storage at 10°C.

The team prepared the mango homogenate from different coated mangoes and determined physicochemical, antioxidant and microbiological changes at different intervals during the shelf life to see the impact of these coating formulations on the packaged mangoes.

'All the treated mangoes had a shelf life of 24 days, whether the essential oils were extracted with methanol or ethanol', says Ayeza Naeem, lead investigator.

At the end of the storage period of 24 days, the firmness of the mangoes decreased, but the sweet aroma of the raw mangoes was intact.

'At the end of the storage period, the colour of the mango seed changed from white to light brown with no morphological changes in the pulp of the fruits. On the other hand, untreated control started decaying after just 8 days of storage', says Naeem.

As the coating is safe for human consumption and prolonged mango shelf life, the National Mango Board can use the research results and popularise the technique among farmers to preserve harvests.

Int. J. Biol. Macromol., **113**: 403–410

Managing Chikungunya Virus
Epitope-based in silico vaccine

The chikungunya virus causes fever and severe joint pain, crippling the infected. Though chikungunya outbreaks have been reported from sixty countries worldwide, there is no commercial vaccine yet for the virus. Dozens of vector-based vaccines are under preclinical testing and two vaccines based on weakened virus particles are under clinical trials.

Scientists from the Central University of Rajasthan have been using *in silico* approaches to design vaccines for malaria, and dengue, as well as for the HIV and Zika virus in their laboratory. Now the team is back with a similar methodology for the chikungunya virus.

Their main focus was the host protein interaction motif, the epitope for vaccine generation. The team predicted such epitopes for all structural and non-structural proteins of the chikungunya virus using prediction servers. And, thus, they homed in on twenty-six sequences with a total of 494 amino acids to design a multi-epitope.

The predicted half-life of the vaccine was 30 hours in human cells.

'The instability index of the vaccine showed that it was stable', says Amit Mishra, IIT Jodhpur.

'The vaccine construct showed a high aliphatic amino acids index, generally correlated to the thermostable property of the proteins', says Aruna Narula, Central University of Rajasthan.

'The epitope sequence is conserved across virus species from different outbreaks', adds Rajan Kumar Pandey.

The team built a structural model for the vaccine, where 10% of amino acids showed disordered regions. They refined the structure model by replacing amino acids with the best possible conformational isomers. Then, using docking studies, they established that the refined epitope structure interacted with human T cell receptors.

'We compared the epitope sequence with known allergen sequences and found no known allergen motifs. So, we expect the vaccine to be non-allergic', says Nazia Khatoon.

'We also cloned the sequence into a vector for vaccine production in *Escherichia coli*', says Vijay Kumar Prajapati.

With industrial vaccine development underway, this *in silico* multi-epitope vaccine could be an alternative to the widely produced weakened virus particle. However, it is essential for all vaccines to follow the development pipeline, including animal and clinical studies.

Infect. Genet. Evol., **61**: 4–15

Oral Insulin
Nanomaterial delivery

Insulin, a peptide hormone, has a significant role in maintaining blood glucose levels. When the body cannot make enough insulin, it is usually injected into the body. If insulin is taken by mouth, it is denatured and broken down by the digestive process. Though there have been attempts to produce formulations to deliver insulin through the oral route, they have not been successful.

Recently, scientists from the Calcutta University and the IIT Roorkee reported developing polyurethane-incorporated chitosan/alginate core-shell nanoparticles for controlled oral insulin delivery.

The scientists prepared chitosan and polyurethane–chitosan nanoparticles and investigated the effects of polyurethane incorporation, within the core or within the shell and within both, using different *in vitro* and *in vivo* parameters.

The team also studied the pharmacological response of insulin-loaded nanoparticles and relative insulin bioavailability after oral administration of insulin-loaded nanoparticles using animal models. They found that bioavailability of insulin ranged from about 9% to about 11% for polyurethane-incorporated chitosan-alginate core-shell nanoparticle formulations – significantly higher than the insulin bioavailability of basic polyurethane core-shell nanoparticle systems.

They also conducted acute toxicity studies for these core-shell nanoparticles using liver function and nephrotoxicity tests in nanoparticle-treated animals and compared the results with those from control animals. 'Polyurethane-incorporated nanoparticles were found to be nontoxic and safe to use as oral drug delivery devices' says P. P. Kundu, IIT Rourkee.

The inclusion of biodegradable polyurethane in the preparation of both core and shell of an oral drug delivery device will definitely unleash a trend of oral insulin delivery. However, more detailed studies are needed to understand how the drug is distributed in the body and metabolised. Animal experiments and clinical trials need to be conducted before diabetics can hope to avoid painful injections.

J. Appl. Polym. Sci., **135**: 46365

Barcoding *Culex* Mosquitoes
Variations in vectors

Since the DNA barcoding of the *Culex* vectors of Japanese encephalitis remains relatively unexplored, researchers from Tamil Nadu, and New Delhi collaborated with scientists in Italy to barcode the DNA of five Japanese encephalitis vectors. They used *Culex fuscocephala*, *Culex gelidus*, *Culex tritaeniorhynchus*, *Culex pseudovishnui* and *Culex vishnui* based on their distribution and vector activity in southern India.

The team took the mitochondrial cytochrome oxidase subunit I gene for barcoding. Mitochondrial DNA is inherited only from mothers and does not undergo recombination from generation to generation. Moreover, there are no introns in the coding of proteins in the mitochondrial DNA of mosquitoes.

The researchers found a conserved region of 648 base pairs that showed marked differences between species. Intra- and inter-species comparisons showed wide divergence and variations. The team calculated intra-species divergence individually for the five species.

Culex tritaeniorhynchus exhibited high polymorphic sites and mutations. The researchers say that this could be due to population size, natural selection, mutation rates, gene flow between populations and introgression from hybridization.

On the other hand, the sister species – *C. vishnui* and *C. pseudovishnui* – showed moderate diversity. The lowest variation was observed for *C. gelidus* and *C. fuscocephala*.

A maximum parsimony tree, created to understand genetic deviations, showed distinct conspecific clusters. The evolutionary tree had seven distinctive clusters, each comprising haplotypes of a given species.

Interestingly, out of the seven clusters, *C. tritaeniorhynchus* occupied three adjacent, but distinctive, subclades based on geographical proximity – Japan, Greece and Turkey and India.

The scientists say that DNA barcoding, based on the cytochrome oxidase subunit I gene, has 100% success rate in identifying mosquito vector species. The DNA barcodes of mosquito species in southern India can help identify species involved in transmitting the virus in a specific region. This, in turn, will help us strategically attack breeding and resting sites, since these sites vary widely between species.

Acta Tropica, **183**: 84–91

PAHs in Indian Food
Causing cancer?

Food habits have changed over the years. Some foods can boost our immune system and others can cause cancer. If, for instance, polycyclic aromatic hydrocarbons – PAHs – contaminate food, they can cause cancer. PAHs are carcinogens and enter the food chain through different routes.

Recently, researchers from the National Institute of Food Technology Entrepreneurship and Management, Haryana, reviewed the magnitude of these compounds in Indian food products. They searched ScienceDirect, Google Scholar, Springer and the Royal Society of Chemistry for research on Indian foods between 1990 and 2017.

The team theoretically evaluated dietary exposure effects to quantify cancer risks. They assessed the risk in different age groups and on urban and rural populations, using deterministic and probabilistic methods.

The scientists observed that children were the most susceptible and that urban, rather than rural, populations had greater risk. The researchers also report that more than fifty per cent of the samples contained these carcinogenic compounds. More than forty per cent exceeded safe limits.

Among food products, the highest concentrations of PAHs were in coconut biscuits. Next came decoctions from cumin seeds, mint leaves and cardamom. Salted, sun-dried and fried chillies also had high PAH content. Low amounts of these compounds were observed in certain food supplements also as well as in *Catla* fish.

The researchers say that environmental pollution and food processing techniques may be factors for increased PAHs in food. PAHs are produced from the incomplete combustion of organic matter.

'They can be dispersed into the environment through industrial and culinary emissions. Processing food and cooking at very high temperatures also produce these compounds', states Tripti Agarwal, NIFTEM, Haryana.

'Baked, roasted and fried foods have high levels of these compounds and their consumption should be reduced', says her colleague, Lochan Singh.

Glen Edelson via Wikimedia Commons

PAHs have been reported to cause skin, bladder, lung, liver and stomach cancer in human beings. The most comprehensive legislation on these compounds in food is by the European Union. They have also set maximum limits for a group of four PAHs. In all other countries there are regulations but maximum limits are rare. This study is, therefore, an eye-opener for policy makers and food technologists.

Chemosphere, **202**: 366–376

Dyeing Coir Products
Effects of lignin and hemicellulose

Coir products have great demand as they are made from natural fibre. Manufacturers apply dyes on coir to make the products more appealing. Coir contains cellulose, lignin and hemicellulose. Materials having 90% of cellulose exhibit good dye absorption. But the effects of lignin and hemicellulose are not clear yet.

Recently, scientists from the University of Calcutta and the National Institute of Research on Jute and Allied Fibre Technology, Kolkata analysed the effects of lignin and hemicellulose on dyeing. They obtained coir fibres and cleaned them in a non-ionic surfactant to remove impurities and waxy coverings. Then they treated the fibres to partially remove lignin, using sodium chlorate and hemicelluloses, using sodium hydroxide.

The researchers washed the treated fibres and dyed them in alkaline and acidic dyes separately. They estimated the constituents in the fibres using Fourier Transform Infra-red spectroscopy and thermal gravimetric analysis. Delignification for an hour caused breaking of some α-ether interlinks of lignin and carbohydrate molecules. The team found that removal of lignin also caused loss of hemicellulose due to their strong bonding.

The scientists analysed the colour uptake using a reflectance spectrophotometer. They observed that the delignified fibres showed reduced acidic dye uptake but improved basic dye uptake. They attributed this to the increased carboxyl content during delignification. They also found that hemicellulose removal resulted in enhanced uptake and penetration of both the dyes. This is due to increased acidity and alpha-cellulose content with structural swelling. The researchers also observed better fastness properties in fibres with lower hemicellulose.

The team says that conventionally coloured coconut fibres show lower uptake of the dye even at the surface. It is also clear, from the study, that the presence of lignin is favourable for acidic dye absorption. This study increases the possibilities of different colouration techniques in lignocellulosics. Now manufacturers can follow these techniques for dyeing coir products.

Ind. Crops Prod., **117**: 20–27

Restoring Carbon Pools
Reclaiming mining site

Land degradation, soil erosion, air pollution and loss of biodiversity are among the many environmental hazards caused by mining. Carbon stocks in the atmosphere, soil, ocean and crust are affected adversely by mining. However, mining is necessary for the growth of the economy. The way out of this impasse is to reclaim mine spoils using vegetation.

Recently, Subodh Kumar Maiti and Jitendra Ahirwal of the IIT Dhanbad reported soil quality improvement and increase in an ecosystem carbon pool at a 16-year-old mine reclamation site. They selected the Rohini opencast coal mining project area of the Central Coalfields Limited, Jharkhand. And used an undisturbed Sal forest nearby as reference site.

In 2001, before the onset of the monsoon, they planted 6-month-old saplings, of eight different drought-resistant multipurpose tree species, on the overburden dump sites of the selected area, to provide economic returns to local communities. They did not apply fertilisers or topsoil, before or after the plantation. In February 2017, the team randomly selected a total of five 10 m × 10 m plots for sampling, in both study and reference sites. They measured abundance, density and plant characteristics in these plots.

The scientists estimated the vegetation survival rate on the reclaimed site to be a little more than sixty per cent of the original, which they attributed to the lack of any treatment. Four tree species had naturally colonised the dump, forming 17% of plant cover. These included the silk cotton tree, the jungle-cork tree, the charcoal-tree and the Indian jujube.

Image: Sanghamitra Deobhanj

The team also found that translocating top soil, during the mining and mixing of spoil materials excavated from different depths, reduced the fine earth fraction in the resulting technosols.

The scientists report that vegetation on reclaimed land improved the quality of technosols. 'The recovery of soil organic carbon was 81% more and soil nutrients, like available nitrogen, were 125% more. Compared to that of reference forest soil, available phosphorus was 160% more and exchangeable potassium was 61% more', says S. K. Maiti. The results show that multi-purpose tree planta-

tion helps degraded ecosystems recuperate.

'The technosols, formed by mine spoil on reclaimed land, facilitate the natural colonization of native tree species and increase ecosystem carbon pools', reports Jitendra Ahirwal.

The team concludes that reclaming mine sites is very important for recovering nutrient content and developing carbon pools in a reconstructed ecosystem. The finding is useful for making surface mining eco-friendly and environmentally sustainable.

Catena, **166**: 114–123

Automatic Emotion Recognition

The human mind has remained a puzzle for ages. From marketing to mental health monitoring, there is a dire need for automatic detection of human emotions in candid environments.

The current facial recognition technologies are not sufficient to decode emotions. They detect emotions but these work only within laboratory conditions. Now, Aparna Mohanty and Rajiv Sahay from the IIT Kharagpur have come up with a solution.

The researchers used a machine learning technique, convolution neural network, a bio-inspired technology based on how neurons take up a signal, process it, and elicit an action in the brain. A convolution neural network acts as a stack of detection filters with every subsequent layer searching for more abstract details than the preceding layer.

The researchers employed a Microsoft Kinect sensor to collect colour and depth information from pictures that depict emotions. They then fed the information to the convolution neural network. And compared details from the filters against an in-built dataset of various emotions.

To validate their technique, Aparna and Rajiv used photographs of Bharatanatyam dancers.

Navarasa or the nine emotions along with hand gestures form the soul of Bharatanatyam. The researchers collected photographs of fourteen individuals each performing a navarasa ten times.

A subset of these photographs formed the dataset for training the network. The team used two other subsets for validation and for testing.

One of the *Navarasas*, namely, *Adbhuta* performed in an actual concert.

By tweaking the number of network layers and the detection capacity of each layer, the researchers could achieve high accuracy in detecting emotions.

To validate the efficiency of their technique in unconstrained environments, they used videos from dance concerts. Despite confounding factors such as make-up, lighting and non-frontal postures, their system accurately recognised emotions from the video frames.

In a world where human computer interaction is on the rise, emotion recognition has become imperative. Once considered a superpower in a fictional universe, automatic emotion recognition is now within our reach.

Pattern Recognition, **79**: 97–113

E-waste Recycling
Occupational hazards

The use of electric and electronic equipment has made our life comfortable. But the challenge is to manage the e-waste generated from discarded equipment. Informal processing of e-waste, as done in developing countries, can affect human health. So monitoring concentrations of toxic components from e-waste in recycling environments is imperative.

Recently, Siby John and team from the Punjab Engineering College reported that barium, copper, lead and

zinc are major pollutants in e-waste dismantling sites in Chandigarh and Ludhiana. The researchers chose these areas based on the trade flow chain and e-waste disposal patterns. They studied the effects of the heavy metals on workers in e-waste recycling sectors there.

'This sector is informal and has primitive recycling and unscientific waste management practices. Workers, here, use bare hands, and do not use face masks or eyeglasses. Poor ventilation compounds the issue', says Siby John.

The researchers analysed the sand, working platform dust and wet wipe samples from face, hands and forearms of the workers for the presence of heavy metals at the e-waste dismantling sites. High concentrations of barium, copper, lead and zinc were observed in the soil and dust samples. Chromium, lead and zinc were observed in high concentrations in dermal samples. Chromium contamination in soil from the recycling sites was about seven times that from the soil of non-recycling sites. The researchers note that the workers are exposed to high levels of chromium and have high carcinogenic risk.

Though India formulated the E-Waste Management Rules in 2016, the present state of affairs is due to lack of strict implementation. Recycling should be done as per government norms and pre-defined e-waste recycling procedures should be adopted. Employers in the e-waste recycling sector need to make workers aware about the hazards and adopt processes as per government norms.

Chemosphere, **203**: 426–433

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