



ISSN: 0975-833X

Available online at <http://www.journalcra.com>

International Journal of Current Research
Vol. 15, Issue, 05, pp.24552-24556, May, 2023
DOI: <https://doi.org/10.24941/ijcr.45228.05.2023>

INTERNATIONAL JOURNAL
OF CURRENT RESEARCH

RESEARCH ARTICLE

IMPACT OF MOISTURE ON YIELD AND STABILITY STUDY OF NEEM OIL, SEED COLLECTED FROM DIFFERENT LOCATIONS OF INDIA

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ARTICLE INFO

Article History:

Received 14th February, 2023

Received in revised form

10th March, 2023

Accepted 16th April, 2023

Published online 15th May, 2023

Key words:

Moisture Content, Stability, Oil Yield,
Neem Seed Kernel, Cold Press.

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Citation: Shri Ram Bharat, Dr. Archana Suyal, Suman Kumar Jha and Dr. Rajeev Saini. 2023. "Impact of moisture on yield and stability study of neem oil, seed collected from different locations of India". *International Journal of Current Research*, 15, (05), 24552-24556.

ABSTRACT

The impact of moisture content on yield and study of stability at 25±2 °C/60 %RH±5 % and 40±2 °C/75 %RH±5 % of mechanically cold pressed neem seed kernel oil collected from four different location was examined. The Matured neem seed was obtained and the initial moisture and oil yield was determined in samples (Bihar (B1nk) 37.0%, Rajasthan (R1nk)34.2%, Tamilnadu (T1nk) 35.2%, Uttar Pradesh (UP1nk) 40.4%) using solvent extraction method in laboratory. The initial data of stability generated including organoleptic, moisture (%), acid value (mgKOH/g), Iodine value (g/ml), saponification value (mgKOH/g) and Azadirachtin content (ppm). The recorded moisture and oil yield was compared with the initial oil content and moisture of the seed kernels for all the four location kernels. As the moisture decrease the yield increase and vice versa. The stability data was generated at both the temperature conditions for 6 month of oil extracted from cold pressing to study effect of temperature and humidity on the quality and stability of stored neem oil. All the test results showed that physicochemical properties of neem oil met the specification limit and the oil was best to use even after 6 months with all its goodness properties intact.

INTRODUCTION

Azadirachtin indica (Neem) belong to Mahogany family, is native of tropical South East Asia and found abundance in India (Okonkwo, 2004). The paramount part of the tree is the oil obtained from the neem seed kernel, which has high concentration of oil reported about 30-45% (Ikasari and Indraswati, 2008). Neem oil is a vegetable oil has active ingredients (limonoids, mono-, di-, sesqui-, and triterpenoids, coumarins, lignins, flavonoids, bitters and other phenols) considered as one of the foremost commercially available product of neem use widely in different regions of the globe in various sector include medicinal, agricultural and cosmetic etc. (Schmutterer, 1995, Kumar et al., 2010). The quality standard of the oil differs according to its method of processing whether mechanically (hot or cold expression) or chemically (solvent extraction) (Puri, 1999). Though premier quality neem oil with a majority of phytoconstituents enrich is obtained via cold pressing. The cold pressing process also ensure any potential residual solvents in the oil that can cause health hazards to the consumer. Many researchers have worked on moisture content effect on oil yield but considering single sample but none have looked impact of moisture in yield and stability data of neem seed collected from different locations.

The stability of a neem oil needs to be tested to find out the effect of various environmental factors such as temperature and humidity on oil quality (Bott. at. el. 2007). The recorded results of stability can be utilized for shelf life prediction, proper condition for storage, and recommended instructions for labelling (Saputri et al 2021). This article therefore, is intended to consider the effect of moisture on yield and collection of stability data of seed of different locations to check quality of neem oil in storage within 6 months.

MATERIALS AND METHODS

Collection of Seeds: The 45 kg fresh fruits sample each was collected from four different location Bihar (B1nk), Rajasthan (R1nk), Uttar Pradesh (UP1nk) and Tamil Nadu (TN1nk) respectively. The seeds were cleaned to remove any foreign material (sticks, unwanted leaves, bad seeds, sand and dirt). 2 kg of seed were kept as such, was decorticated and its moisture and oil yield was examined. The 40 kg seeds were air dried and then decorticated using decorticated machine to get neem kernel. The finally air dried kernel were stored in gunny bags in cool, dry, shady, aerated space prior to use.

Instruments and chemicals: All instruments used during study were well calibrated and the chemicals used during study procured from Merck, Germany and Sigma Aldrich, United States.

Moisture Content Determination: The ASAE 1998 standard reference was used to determine moisture content of the seed kernel. The sample was divided into two parts; one part (2kg) was left as it was; while the remaining (40 kg) part was air dried for 48 hours to further reduce the moisture content. The 5g seed sample each from different location were placed in an oven set at 105°C for 3 hours in laboratory. The samples were then cooled in desiccator, weighed and the moisture content calculated. Loss in weight is assumed to be moisture loss. Initial moisture content of the seeds was 12.6, 18.5, 10.4 and 8.7% for B1nk, R1nk, UP1nk and TN1nk respectively. On air drying kernel for 48 hours, the moisture was analysed and was recorded in B1nk, R1nk, UP1nk, and TN1nk was 2.8%, 4.5%, 4.8%, and 3.5% respectively. Three replicates of the moisture experiments were done.

Extraction

Soxhlet Extraction: In this process ground sample of neem seed 50 g (as such) from 2kg was placed in a “thimble” made of filter paper and placed in the Soxhlet extractor. A flask (500 ml) containing 350 ml of the appropriate solvent was attached at the bottom and heated until evaporation, the method was performed according to the 936.15 AOAC (1990) procedure. Each extraction was carried out for 6 hours. Dried in rotatory evaporator and oil content was recorded.

Cold Pressed: The extraction process is based on blend of two methods extrusion and dissolution. The method allows separation of components without thermal degradation of seed content and favour direct extraction of a rich Azadirachtin extract and increasing concentration of other compounds of the neem seed kernels (NIIR Board, 2004). A mechanical expeller with vertical action equipped with a steel cylinder shaped container that can holds upto 12 kg of dehulled seed, a manually operated hydraulic press system (20 kg cm⁻²), and a cylinder with a reduction in the lower exit and a steel drain used to collect the extracted oil at the bottom. The neem kernel each of 10 kg was loaded each and oil was collected separately. According to Olajide (2000) the oil was collected and left to settle down for 96 hours, thereafter volume was measured. The weights of the neem cakes (leftover of kernel after oil extraction) were determined. The expeller oil yield was compared with the initial soxhlet extracted oil yield.

Physicochemical properties of neem oil for stability: The study of stability at two temperature and humidity condition of neem oil through physicochemical properties including organoleptic, moisture (%), acid value (mgKOH/g), Iodine value (g/ml), saponification value (mgKOH/g) and Azadirachtin content (ppm) for ensure its purity over 6 months. Physicochemical stability testing was carried out on neem oil at room temperature (25±2 °C/60 %RH±5 %) and high temperature (40±2 °C/75 %RH±5 %) for 6 month. The test was carried out in triplicate at each storage temperature for all the four samples.

RESULTS AND DISCUSSION

The result showed that neem kernels from all the four location, yield less oil when moisture content is higher compared to those with lower moisture content kernel; this accord with the study by Orhevba *et al.* 2013 who reported maximum oil extracted of 24.86% from neem kernel having moisture content of 8.1%. Further they examine that increasing the moisture content to 16.6% decreased the oil recovery by 15.62%. However, from the figure 1, the effect of moisture on yield of neem oil can easily be understood. As the initial moisture was examine the moisture level was high in kernel so the oil yield (a) was low and as the moisture content decrease due to air drying for 48 hours the oil yield

increase in all the location examined. The initial result of moisture content was recorded 12.6, 18.5, 10.4 and 8.7% and oil yield was 24.2, 15.4, 27.44 and 30.6 % in B1nk, R1nk UP1nk and TN1nk respectively. While when moisture content decrease 2.8% in B1nk, oil yield was 40.01%, 4.5% in R1nk, oil yield was 38.4%, 4.8% in UP1nk, oil yield was 41.7% and 3.5% in TN1nk, oil yield was 36.6%. The result of the physicochemical properties determination was compared to the specification of IS 4765 – 1975 certificate of analysis. The moisture, acid value, Iodine value, saponification value and Azadirachtin content of neem oil as per specification should be NMT 1%, NMT 15 mgKOH/g, limit 65-80 (g/ml), limit 180-205 (mgKOH/g) and as such respectively. The organoleptic characteristic was performed by visually observing the colour and odour of the neem oil. Moisture was done using Karl Fisher auto Titrator. The acid value, iodine value and saponification value was done using titration method. The Azadirachtin content was analysed using HPLC. Physicochemical test of neem oil considering ambient temperature (25±2 °C/60 %RH±5 %) and high temperature (40±2 °C/75 %RH±5 %) for all the four location is shown in figure 2a and 2b. The study shows that all physicochemical properties of neem oil used in this study met the specification followed.

Organoleptic is used for verifying oil physical appearance. Acid value is used to calculate the quantity of free fatty acids present in oil or fat. If the oil have excessive fatty acid content, it will speed up the process of rancidity, which will decrease quality of oil received. Iodine value represents the degree of unsaturation of oil or fat. The greater iodine value that means higher degree of unsaturation and quality of oil is exorbitant. Saponification value define the size of the fat molecule. The greater saponification value of the fat or oil, the size of the molecule is smaller. In both the temperature and humidity condition organoleptic was found to be stable from initial to 6 month that was greenish brown to dark brown colour with characteristic odour. The moisture content derive from air humidity keep on slowly interacting with oil molecules and though moisture kept increasing overtime in both the stability conditions in all the four samples. The sample of UP1nk show the high moisture in both the temperature condition above all sample analysed during 6 months. The low acid value is an indication of good non-degraded state of oil, and was within the limit in sample tested in stability. It kept rising in all stability samples but it shown fast increase in case of high temperature (40±2 °C/75 %RH±5 %) and slow increase in case of ambient temperature (25±2 °C/60 %RH±5 %). The neem kernel of R1nk was recorded high acid value in 6 month in both temperature condition. The iodine value show high unsaturation of the oil examined in all four samples show variation from low to high and vice versa in both the stability temperature, but the high fluctuation was seen in high temperature (40±2 °C/75 %RH±5 %) due to the fact that heating highly unsaturated fatty acids results in polymerisation of glycerides which could lead to the formation of deposits.

The saponification value was high during initial testing but decrease in middle months and again increase in 6 month. The saponification value of oil extracted was determined and found to be in the range of 190.14 – 204.65 at 25±2 °C/75 %RH±5 % and range from 187.18 – 204.65 at 40±2 °C/75 %RH±5 %. The high saponification value reported in both condition showed that the oil is normal triglyceride and it is highly beneficial in the production of soap and shampoo (Akbar *et al.* 2009). Azadirachtin initial content was recorded from highest to lowest in TN1nk, R1nk, B1nk and UP1nk (Figure 3). Instability sample active ingredient level continuously showed degradation in both storage condition from 3400 – 870 at 25±2 °C/75 %RH±5 % and 3400 to 751 ppm at 40±2 °C/75 %RH±5 % respectively. As per Saputri *et al.* 2021, the Azadirachtin level in neem oil which was used in cream showed degradation within 1 month at both the storage temperature conditions. Cream made of neem oil was stable for 1 month at 25 °C and for 1 week at 40 °C. Which shows Azadirachtin degrades fast at high temperature condition in

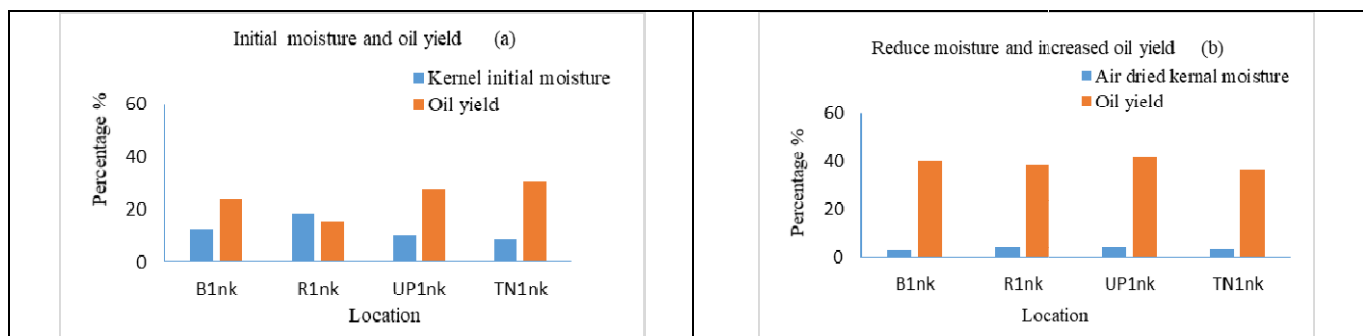


Figure 1: (a and b) Variation of oil yield due to moisture content

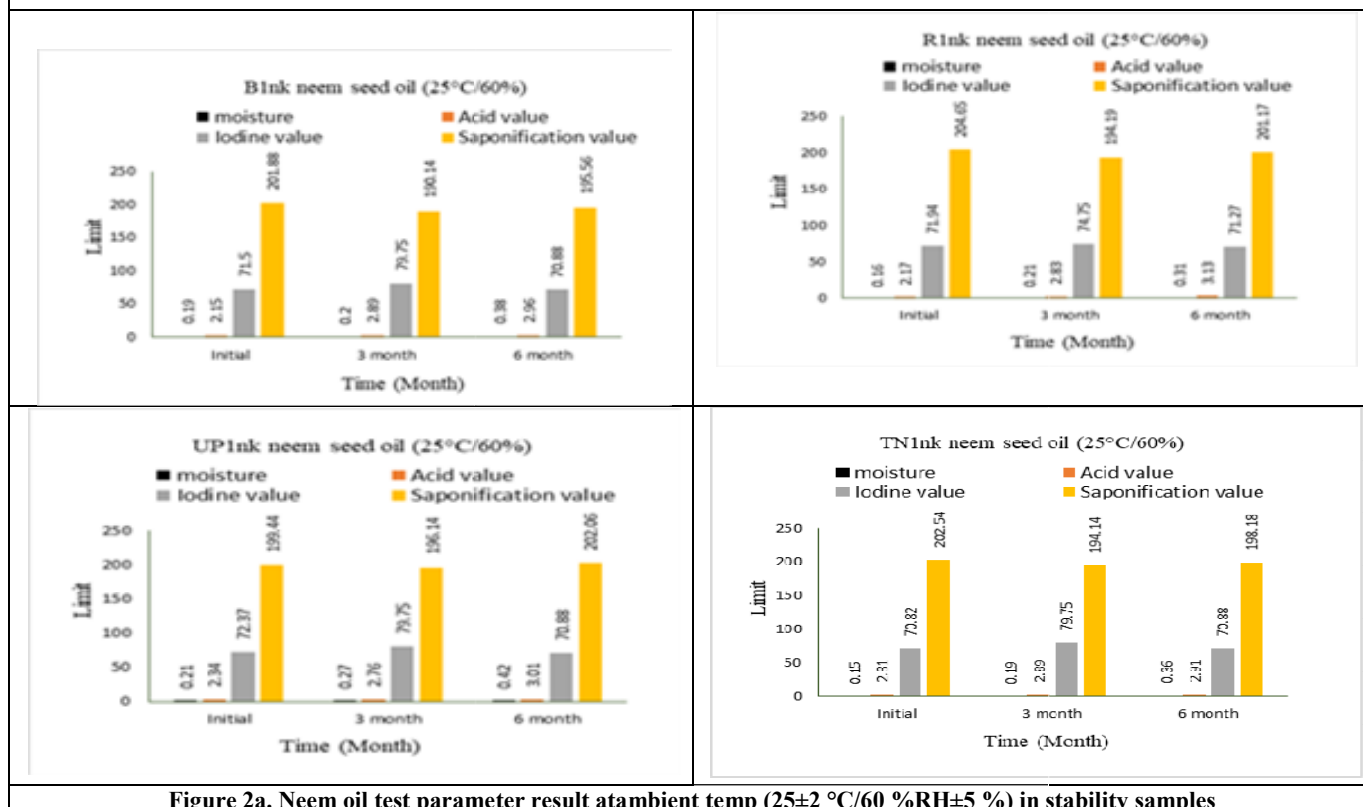


Figure 2a. Neem oil test parameter result at ambient temp (25±2 °C/60 %RH±5 %) in stability samples

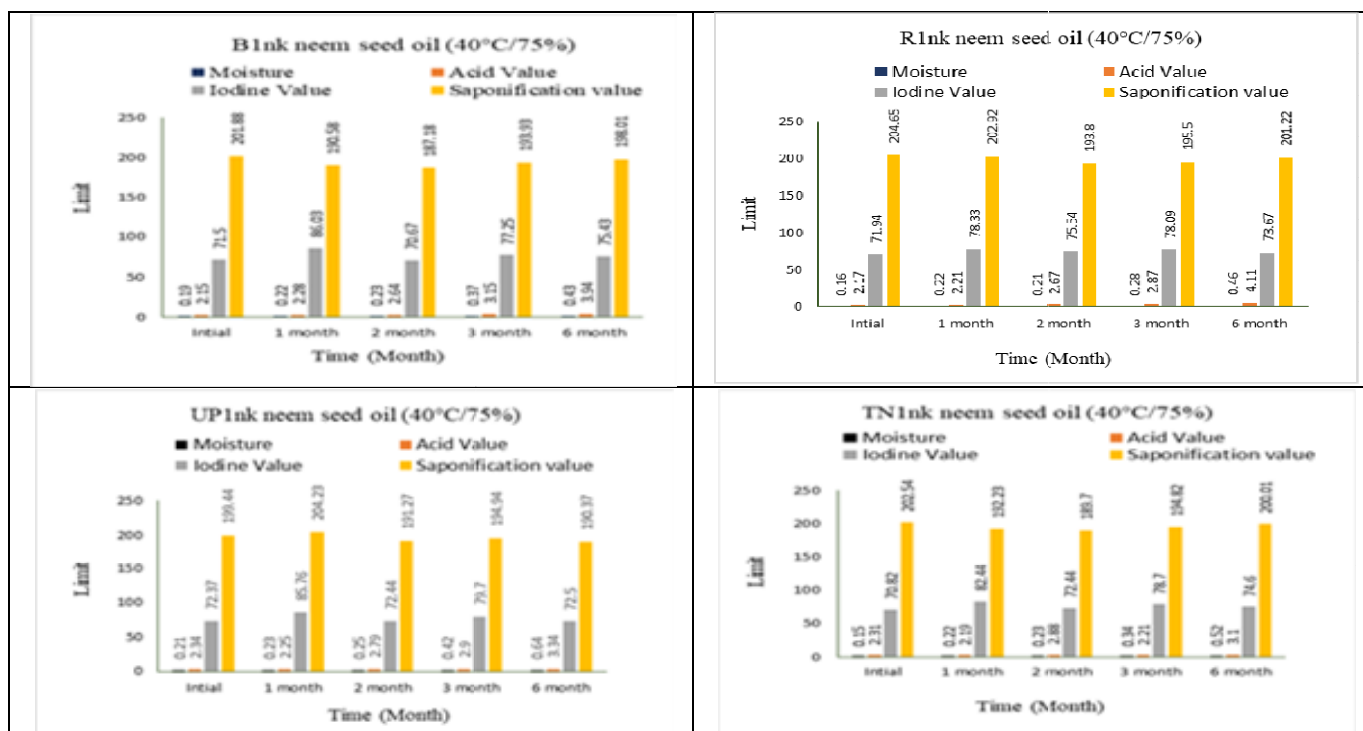


Figure 2b. Neem oil test parameter result at high temp (40±2 °C/75 %RH±5 %) in stability samples

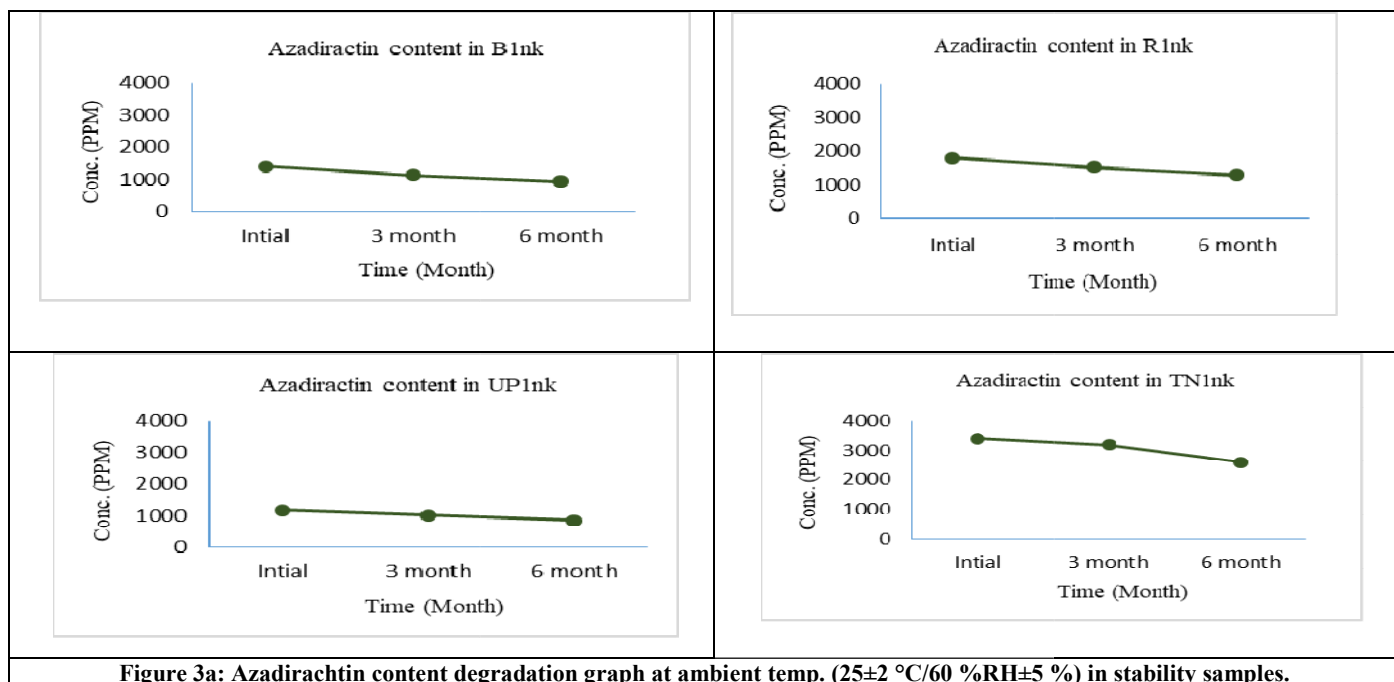


Figure 3a: Azadirachtin content degradation graph at ambient temp. ($25\pm 2\text{ }^{\circ}\text{C}/60\text{ \%RH}\pm 5\text{ \%}$) in stability samples.

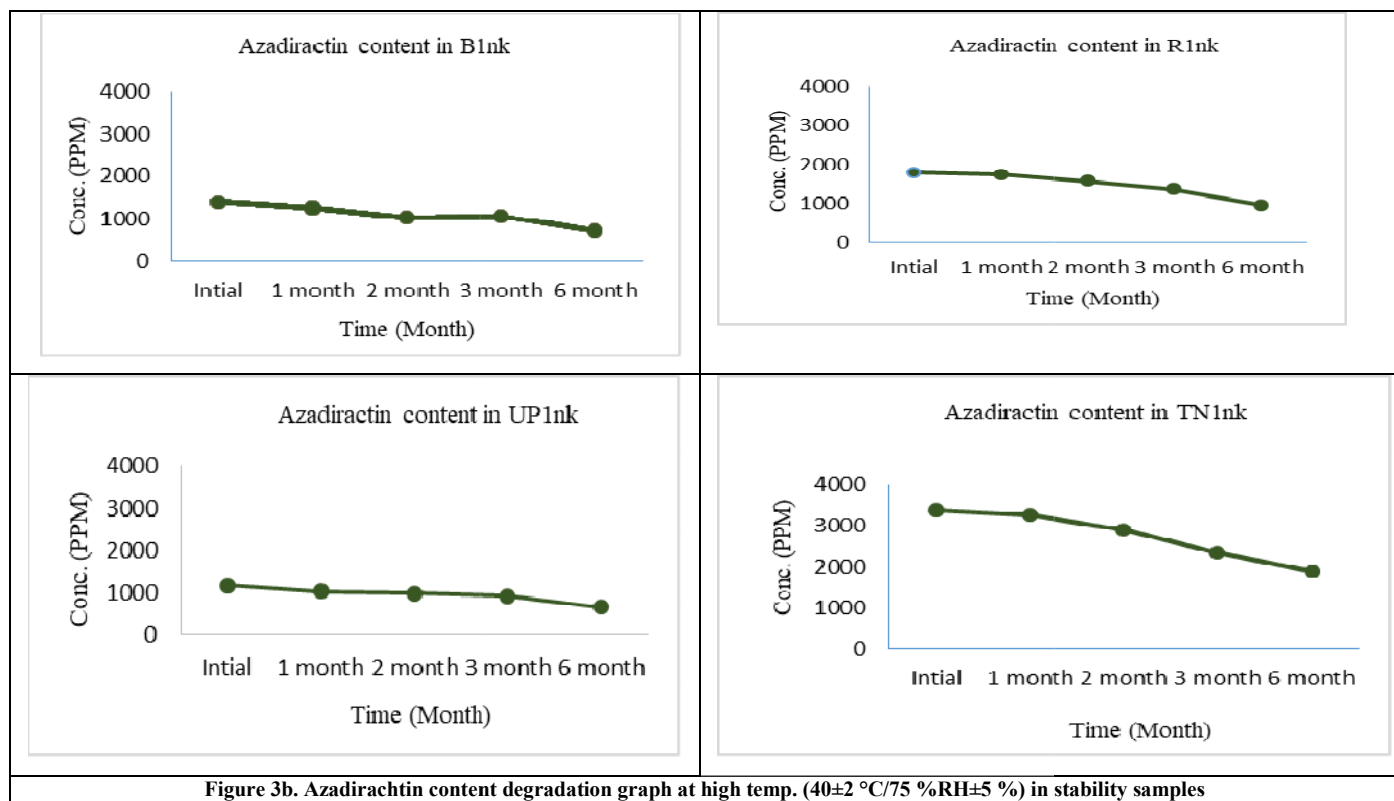


Figure 3b. Azadirachtin content degradation graph at high temp. ($40\pm 2\text{ }^{\circ}\text{C}/75\text{ \%RH}\pm 5\text{ \%}$) in stability samples

comparison to ambient temperature. The Azadirachtin degradation curve is shown at $25\pm 2\text{ }^{\circ}\text{C}/75\text{ \%RH}\pm 5\text{ \%}$ and $40\pm 2\text{ }^{\circ}\text{C}/75\text{ \%RH}\pm 5\text{ \%}$ in figure 3a and 3b respectively. The observation result of the tests carried either at room temperature ($25\pm 2\text{ }^{\circ}\text{C}/60\text{ \%RH}\pm 5\text{ \%}$) and high temperature ($40\pm 2\text{ }^{\circ}\text{C}/75\text{ \%RH}\pm 5\text{ \%}$) showed that neem oil was stable in organoleptic and homogeneity for 6 months of storage.

CONCLUSION

The study shows the moisture play key role in percentage of oil yield, as the moisture content increase the oil yield reduces and vice versa in neem kernel collected from all the four location (Bihar (B1nk), Rajasthan (R1nk), Tamilnadu (T1nk), Uttar Pradesh (UP1nk)).

Further, all the physiochemical parameter tested in neem oil within 6 months at two conditions i.e. ambient temperature ($25\pm 2\text{ }^{\circ}\text{C}/60\text{ \%RH}\pm 5\text{ \%}$) and high temperature ($40\pm 2\text{ }^{\circ}\text{C}/75\text{ \%RH}\pm 5\text{ \%}$) was recorded seems to be in limits. However, the Azadirachtin content shows degradation in both storage condition whether ambient or high temperature. Further, rapid degradation was recorded at high temperature in comparison to ambient temperature of storage. The high temperature condition altered the chemistry of Azadirachtin content due to which it showed fast break down. Meanwhile, all the test results showed that physicochemical properties of neem oil met the literature specification limit in both the storage condition. The cold press neem oil was up to the mark even after 6 months with all its goodness properties fully intact.

ACKNOWLEDGEMENTS

The author's would like to acknowledge Patanjali Ayurved Limited and Patanjali Foods Limited, Padartha, Haridwar for providing all necessary details in preparation of this manuscript.

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