SYNTHESIS, CHARACTERIZATION AND OPTIMIZATION STUDY OF NICOTINE DERIVATIVES

Yogesh Baj[1], Savankumar Patel[2], Nirmal K. Patel[3]
[1,2,3]Department of Industrial Chemistry, Institute of Science & Technology for Advanced Studies & Research, Sardar Patel University, V. V. Nagar – 388120, Gujarat, (INDIA)

Abstract: Oxidation of nicotine leads to synthesis of nicotinic acid, Nicotinic acid also known as pyridine-3-carboxylic acid or niacin, a water soluble B-complex vitamin found in various plants and animals and has vital roles in biological processes as production of energy, signal transduction, regulation of gene expression and synthesis of fatty acids, cholesterol and steroids, while the deficiency of nicotinic acid in man leads to complex pathological condition, known as pellagra. Tobacco is main source of nicotine and approximately 0.6% to 8.0% of nicotine is present in various part of tobacco plant. In present study extraction of nicotine from tobacco waste was carried out, further the extracted nicotine was subjected to oxidation for the synthesis of nicotinic acid by using hydrogen peroxide. Reaction parameters such as temperature, time, volume and concentration of toluene and hydrogen peroxide were optimized. In result, 1.2 g of nicotine was extracted from tobacco waste at 60^0C in 150ml toluene in 6 h. Whereas 3.87 g of nicotinic acid was procured by using 50 ml, 2M solution of hydrogen peroxide in 8 h time at temperature 70 ^{0}C. Extracted nicotine and nicotinic acid was characterized by mass spectroscopy and fourier transforms infrared spectroscopy respectively.

Keywords: extraction, nicotine, nicotinic acid, niacin, vitamin, oxidation.

I. INTRODUCTION

Numbers of organic compound which were found to be essential in small amounts for the normal physiological functions of higher animals and were not synthesized in their bodies were collectively designated as vitamins [1].

In chemical composition and properties the vitamin varies widely among themselves. They constitute, therefore, a heterogeneous assemblage forming like the hormones only a biologically defined group Deficiency of vitamins in the diet cause various pathological conditions most of which can be reversed by their proper administration[2]

Nicotinic acid (pyridine-3-carboxylic acid), also known as niacin which is water soluble B-complex vitamin found in various plants and animals and has vital roles in biological processes as production of energy, signal transduction, regulation of gene expression and synthesis of fatty acids, cholesterol and steroids[3]. While the deficiency of nicotinic acid in man leads to complex pathological condition, known as pellagra the main symptoms of the disease include inflammatory changes of skin and lead to mental desperation [4]

It is also used in the treatment of Atherosclerosis because it is a hypolipidemic agent and also used in the treatment of multiple sclerosis [5]. The exact mechanism of action of nicotinic acid in multiple sclerosis is not known. Some evidences suggest that it dilates the blood vessels of the brain and provides more oxygen to the brain and reduces the symptoms [6]

Nicotinic acid can also be chemically modified to various derivatives as it consist of pyridine nucleus is an important hetero-aromatic class of compounds with a wide range of activities [7]. Nicotinic acid derivatives and its isomers have also been investigated as an agent for the prevention or delay of the onset of type 1 diabetes mellitus [8]. They also have anti-bacterial, anti-oxidant, anti-inflammatory and anti-carcinogenic activities, and have putative activity against osteoarthritis and granuloma annular. For example, it can be mentioned the importance of anti-tuberculosis first line drug Isoniazid [9], which is an analogue of iso-nicotinic acid, an isomer of nicotinic acid. Nicotinic acid derivatives are also an important start material for the preparation of other biological activity compounds. Considering that, 2-chloro-3-pyridine carbonyl chloride is a useful intermediate for preparation of Nevirapine (Viramune), a valuable anti-AIDS drug [10]. Due to this importance of nicotinic acid derivatives and its isomers have significant role in pharmaceutical industries

The nicotinic acid was first isolated from rice polishing and later on it was also isolated from yeast [11]. As it is an oxidation of nicotine, while tobacco is main source of nicotine approximately 0.6% to 8.0% of nicotine is present in various part of tobacco plant So an integrated approach for isolation of nicotine from tobacco waste has been proposed [12] As rational production and processing of tobacco plant must include the entire biomass, both the main product leaves as well as stalks that remain after harvest. Residues (stalks and small leaves) and significant amounts of leaf scrap and waste generated during processing of tobacco, can serve as a very important secondary raw material for nicotine, by final processing, nicotine acid could be obtained in industry which can be useful as various pharmaceutical agent [13].

Based on these considerations, the present work investigates the extraction of nicotine from tobacco wastes & they are chemically modified to their derivative known as nicotinic acid. Extracted nicotine and prepared derivative of nicotine was characterized by Mass spectroscopy (M S) and Fourier transform infrared spectroscopy (FTIR) respectively.

II. Experimental

Tobacco waste was obtained from Mahi tobacco. Calcium hydroxide, toluene hydrogen peroxide, dimethyl ether, ethanol were purchased from Atul Chemical, Anand. Solvents and other laboratory chemical were used after routine purification and they were of analytical reagent grade. Double distilled water was used.

III. Methods

Extraction of nicotine

Accurately weigh 20 g of sample in thimble & put in soxhlet assembly with round bottom flask, Add 150 ml of toluene as solvent. Now put assembly in heating mental and carried out extraction for refluxing temperature for 6 hr. After completion of extraction mixture of nicotine and toluene is separate using simple distillation.

Preparation of nicotinic acid

The above extracted nicotine was further used for preparation of nicotinic acid which is also known as niacin which was prepaid by oxidization of extracted nicotine by using hydrogen peroxide as oxidizing agent. 2 moles of hydrogen peroxide, 1 mole of nicotine was added to round bottom flask equipped with stirrer. The reaction mass was heated for several hours, then cool down to room temperature. Absolute dimethyl ether was added to the reaction mixture which effect precipitation of crystals. The crystals were filtered, dried and recrystallized in absolute ethanol.

Mass Spectroscopy

Mass Spectroscopy is a technique used to measure the characteristic of a sample or molecule i.e., chemical composition and structure. This will be happened by converting the material to charged molecules to measure their mass to charge ratio. All mass spectrometry techniques have a similar component, but these components sometimes differ in their nature from one mass spectrometry to another one depend on the type of mass spectrometry but all the mass spectrometry machines work with a similar way, the following three steps show how this technique works:

• Ionization of the small sample by losing an electron (this called the ionizer).
• The ions will be sorted and separated according to their mass and charge (this called the analyzer).
• Then the separated ions then measured and the data will be displayed on a chart (this called the detector). The resulting products were characterized by mass spectroscopy using LCQ Fleet Ion Trap LC/MS instrument by the solvent dissolving method.

FTIR Spectroscopy

Fourier transform infrared spectroscopy (FTIR) is a technique which is used to obtain an infrared spectrum of absorption, emission, photoconductivity or Raman scattering of a solid, liquid or gas. An FTIR spectrometer simultaneously collects high spectral resolution data over a wide spectral range. This confers a significant advantage over a dispersive spectrometer which measures intensity over a narrow range of wavelengths at a time. In infrared spectroscopy, IR radiation is passed through a sample. Some of the infrared radiation is absorbed by the sample and some of it is passed through (transmitted). The resulting spectrum represents the molecular absorption and transmission, creating a molecular fingerprint of the sample. Like a fingerprint no two unique molecular structures produce the same infrared spectrum. This makes infrared spectroscopy useful over several types of analysis. The resulting products were characterized by FTIR spectroscopy using Perkin Elmer spectrum GX instrument, by the KBr pallet method.

IV. RESULT & DISCUSSION

Extraction of nicotine

The extraction of nicotine from tobacco waste was carried out successfully by solvent extraction method by using sohxlet assembly; the extracted nicotine was evaluated by its physical properties such as color, boiling point and solubility. The nicotine obtain was yellow-brown/oily liquid that is readily soluble in alcohol, it was miscible in water and the boiling point obtain was 240^0C, which conform that extracted martial was nicotine.

Optimization of nicotine extraction was carried by varying amount of solvent (toluene), temperature and time of reaction which is shown in below mention tables respectively.

Table 1. Effect of solvent

No.	Solvent (ml)	Temperature (^{0}C)	Time(h)	Result (nicotine in g)
1	50	60	6	0.85
2	100	60	6	1.00
3	150	60	6	1.20
4	175	60	6	1.20

In the above table 1, amount of solvent is varied from 50 ml to 175 ml by keeping time and temperature constant at 6 h and 60 ^{0}C, respectively. From above table one can conclude that optimum amount of solvent required for extraction of nicotine is 150 ml. Also the volume of solvent has a significant effect on extraction efficiency shown in the table; it is also observe that as the volume

of solvent increases the amount of extracted nicotine increases. The possible reason for the increase efficiency with the increasing amount of the solvent might be due to the increase in swelling of tobacco waste material by solvent, which increase the contact surface area between the tobacco waste and solvent. So the extraction of nicotine compound from the tobacco waste material is directly related to the amount of solvent.

Table 2. Effect of temperature

No.	Solvent (ml)	Temperature (^{0}C)	Time(h)	Result (nicotinic acid in g)
1	150	70	6	0.92
2	150	65	6	1.09
3	150	60	6	1.2
4	150	6	6	1.45

In the above table 2, temperature was varied by keeping amount of solvent and reaction time constant. The temperature was varied from 55 ^{0}C to 70 ^{0}C at interval of 5 ^{0}C but best result was obtained at 60 ^{0}C. The extraction temperature was another important influencing factor in the extraction of nicotine. The above table indicates that a significant increase in extraction of nicotine with increase in temperature. This is due to increase solubility and diffusion coefficients of nicotine; decreased solvent viscosity; as well as enhanced mass transfer and penetration of solvent in to the tobacco waste, thus accelerating the whole extraction on other hand heating might soften the tobacco waste tissue, so nicotine might be easily extracted, so the moderate temperature of extraction is 60 ^{0}C but at higher temperature 70 ^{0}C the yield was lower because solvent might be started evaporating which effects directly the loss of solvent to solid ratio.

Table 3. Effect of time

No.	Solvent(ml)	Temperature(^{0}C)	Time(h)	Result (nicotine in g)
1	150	60	2	0.79
2	150	60	4	0.92
3	150	60	6	1.20
4	150	60	8	0.95

The above table 3 shows that reaction time was varied by keeping amount of solvent and temperature constant. Reaction time varies from 2 h to 8 h at interval of 2 h. From the obtained data it is clear that the optimum reaction time is 6 h. The selection of an appropriate extraction time was the final step in a series of single factor experiments. The above table shows that nicotine extracted increased gradually with increase in time, these phenomena could be explained by the fick's second law of diffusion, predicting that a final equilibrium between the solute concentration in the solid matrix and in the solvent might be reacted after certain time, leading deceleration in the extraction yield. Moreover prolong extraction time increase the chance of decomposition of desired compound. So the moderate time was 6h.

So it can be conclude that optimum temperature was 60 ^{0}C with 6 h of extraction time by using 150 ml of chloroform as extracting solvent for fruitful extraction of nicotine from tobacco waste and resulting nicotine was confirmed by mass Spectroscopy which is represented below.

Mass Spectra of Nicotine

The Figure 1 is the mass spectrum of Nicotine in 1+VE. The molecular formula of Nicotine in $C_{10}H_{14}N_2$ having molecular weight 162.24. Here the base peak with 100% relative intensity which corresponds to the most abundant ion is 163.06 can be of our target molecule i.e. nicotine. As per rules of mass spectroscopy here M+1 (13%)/(1.1%) abundance of carbon isotope = 11.8 shows presence of 10 to 11 carbon atoms and M+2 (4%)/(4.29%) ^{34}S isotopic mass of sulphur shows there is no presence of sulphur atom. AMU left over for other atoms = M – C_{10} – N_2 = 163.06 – 120 – 28 = 15, which is equal to the summation of total hydrogen atom in the structure. So from this it can be said that the below mass spectra is of nicotine.

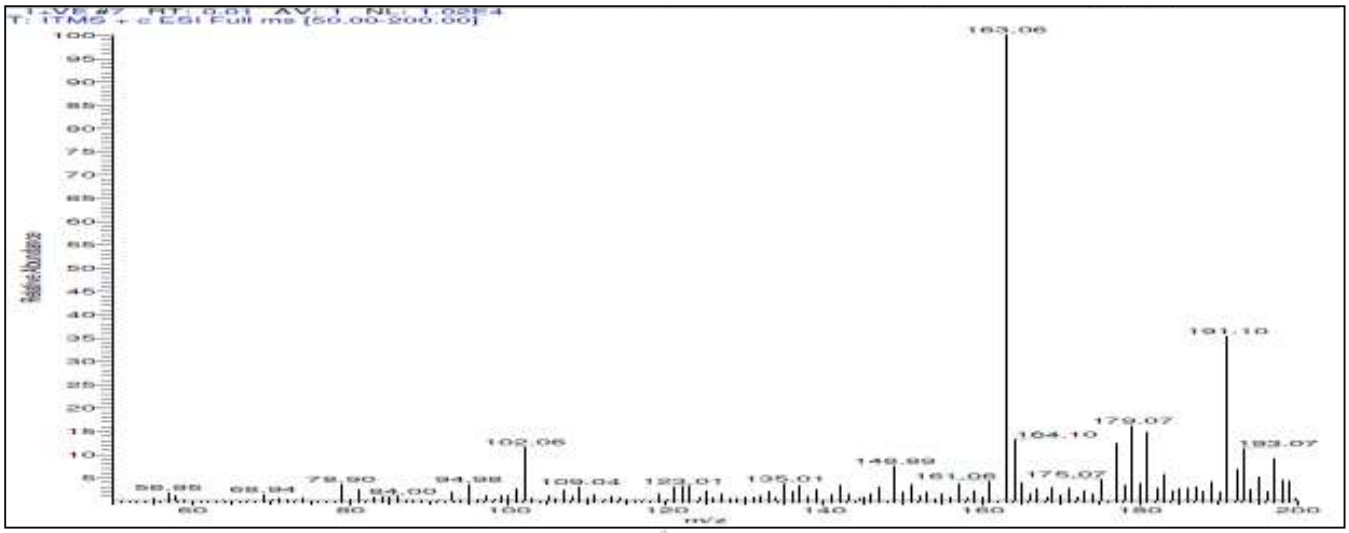

Fig.1

Oxidation of nicotine

The hydrogen peroxide is one of the oxidizing agents for successful oxidation of nicotine, it has been conveniently used in reaction below 100^0C, and the time of reaction depends largely on temperature so Optimization of nicotine oxidation was carried by varying amount of solvent concentration, temperature and time of reaction which is shown in below mention tables respectively. about 6 h at 70^0C nicotine acid formed during the reaction was evaluated physical properties such as color, melting point and solubility, nicotinic acid obtain was off-white soiled, with melting point 230^0C that was readily soluble in water, which may confirm that synthesized martial was nicotinic acid.

Table 4. Effect of concentration of hydrogen peroxid

No	Concentration of hydrogen peoxide (M)	Time(h)	Temperature (^{0}C)	Result(nicotine acid obtains in g)
1	0.5	8	70	1.25
2	1.0	8	70	1.87
3	1.5	8	70	2.15
4	2.0	8	70	3.85

Above table 4 states that concentration of hydrogen peroxide varied from 0.5 M to 2.0 M, time and temperature was kept constant. Maximum yield was obtained at 2.0 M because reactions proceed faster at higher concentrations as more reactant molecules are available and therefore collisions between reactant molecules are more likely. Here reaction between nicotine and hydrogen peroxide is carried out .When a dilute hydrogen peroxide is added to nicotine; a pale white precipitate of nicotinic acid was formed. As the nicotine were diluted more and more, so the precipitation also took longer time to form, as the concentration of hydrogen peroxide increased precipitation also started earlier with good yield.

In order for any reaction to happen, those particles must first collide. This might true whether both particles are in solution, or whether one is in solution and the other a solid. If the concentration is higher, the chances of collision are greater.

Table 5. Effect of time

Serial no	Concentration of nitric acid(M)	Time (h)	Temperature (^{0}c)	Result (nicotinic acid obtain in g)
1	2.0	4	70	1.65
2	2.0	6	70	2.35
3	2.0	8	70	3.87
4	2.0	10	70	3.87

Above table 5, show the effect of time on reaction medium. The reaction time was varied by keeping amount of solvent and temperature constant. Reaction time was varied from 2 hto 8 h with 2 h interval. The optimum reaction time was 8 h may be due to extended time allows the reactant to react sufficiently to form the product.

Table 6. Effect of temperature

No.	Nitric acid(2.0 M) solution in (ml)	Temperature (^{0}C)	Time(h)	Result (nicotinic acid in g)
1	50	75	8	1.92
2	50	70	8	1.92
3	50	65	8	1.09
4	50	60	8	1.2
5	50	55	8	0.45

Above table 6 states varied reaction temperature from 55^{0}C to 70 ^{0}Cand amount of solvent with 2 molarity was kept constant. Maximum yield was obtained at 70^{0}C. The oxidation rate increases significantly with the temperature. It can be seen that at 70^{0}c the yield is higher, if the temperature is decreased from 70^{0}c to 55^{0}c yield also decreased. The main reason of this striking temperature influence on the oxidation rate is the temperature dependence of the diffusivity of oxygen (O_2) atom of hydrogen peroxide (H_2O_2) into nicotine ($C_{10}H_{14}N_2$). The diffusivity of the oxidants depends on the temperature T. The oxidant diffusivity is exponentially increased with higher temperature and exponentially decreased with lower temperature. Higher diffusivity means that more oxidants interface and react there with nicotine ($C_{10}H_{14}N_2$) to nicotinic acid ($C_6H_5NO_2$).

So it can be conclude that using 2 molar hydrogen peroxide as oxidizing agent can convert nicotine to nicotinic acid with temperature 70 ^{0}C and 8 h of reaction time, obtain nicotinic acid was confirmed by FTIR which is mention below.

FT-IR spectra of Nicotinic acid

Figure 2 show the FT-IR spectrum of the Nicotinic acid. Here the frequency with strong intensity at 3440.68 cm^{-1} belongs to carboxylic acid –OH stretching, 3072.02 cm^{-1}belongs to weak =C-H stretch and the weak band at 1485.20 is of aromatic C=C which confirm the structure that is nicotinic acid.

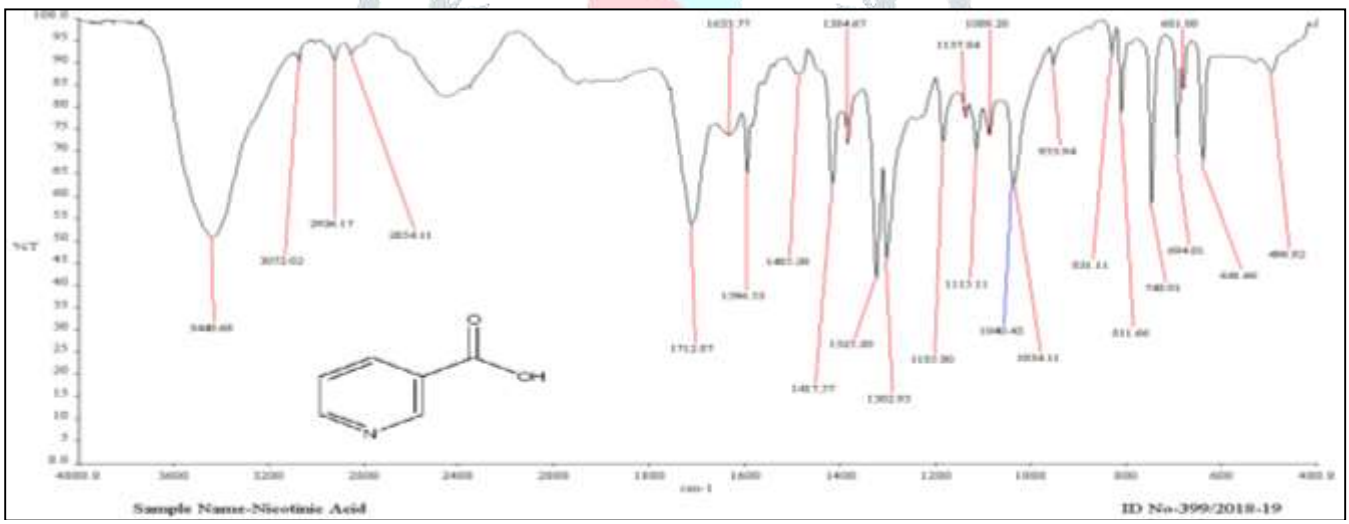

Fig. 2

V. CONCLUSION

Nicotine was successfully extracted from tobacco waste by solvent extraction methods using soxhlet apparatus. Fruitful extraction was carried out and the extracted compound was confirmed by mass spectroscopy. Further nicotinic acid was prepared fruitfully by oxidation of extracted nicotine using hydrogen peroxide as oxidation agent and the structure was confirmed by FTIR. Reaction parameters such as temperature, time, volume and concentration of toluene and hydrogen peroxide were optimized. In result of nicotine was extracted from tobacco waste at 60 ^{0}C in 150 ml toluene in 6 h while nicotinic acid was successfully obtain by oxidation using 50 ml, 2M solution of hydrogen peroxide in 8 h at temperature 70 ^{0}C, further nicotinic acid can also be chemically modified to various derivatives as it consist of pyridine nucleus which stands in hetero-aromatic class of compounds with a wide range of pharmaceutical activities

REFERENCES

[1].Hubalek, V., Buck, M., Tan, B., Foght, J., Wendeberg, A., Berry, D., Bertilsson, S. and Eiler, A. 2017. Vitamin and amino acid auxotrophy in anaerobic consortia operating under methanogenic conditions. Journal of *MSystems*, *2*(5):38-17.

[2].Stevens, G.A., Bennett, J.E., Hennocq, Q., Lu, Y., De-Regil, L.M., Rogers, L., Danaei, G., Li, G., White, R.A., Flaxman, S.R. and Oehrle, S.P. 2015. Trends and mortality effects of vitamin A deficiency in children in 138 low-income and middle-income

countries between 1991 and 2013: a pooled analysis of population-based surveys. *The Journal Lancet Global Health*, *3*(9):528-536.

[3].Devalia, V., Hamilton, M.S. and Molloy, A.M. 2014. Guidelines for the diagnosis and treatment of cobalamin and folate disorders. *British journal of haematology*, *166*(4):496-513.

[4].Wang, W. and Liang, B.2012. Case report of mental disorder induced by niacin deficiency. *Shanghai archives of psychiatry*, *24*(6):352.

[5].Eckel, R.H., Jakicic, J.M., Ard, J.D., De Jesus, J.M., Miller, N.H., Hubbard, V.S., Lee, I.M., Lichtenstein, A.H., Loria, C.M., Millen, B.E. and Nonas, C.A. 2014. 2013 AHA/ACC guideline on lifestyle management to reduce cardiovascular risk: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. *Journal of the American College of Cardiology*, *63*(25 Part B):2960-2984.

[6].Pan, L., Yu, G., Chen, X. and2 Li, X .2017. Nicotinic acid inhibits angiogenesis likely through cytoskeleton remodeling. Journal of *Organogenesis*, *13*(4):83-191.

[7].Saxena, S. and Jha, S. 2017. Role of NOD-like Receptors in Glioma Angiogenesis: Insights into future therapeutic interventions. *Cytokine & growth factor reviews*, *34* :15-26.

[8].McElvain, S.M. and Goese, M.A. 1941. The preparation of nicotinic acid from pyridine. *Journal of the American Chemical Society*, *63*(8) :2283-2284.

[9].Itzhaki, O., Greenberg, E., Shalmon, B., Kubi, A., Treves, A.J., Shapira-Frommer, R., Avivi, C., Ortenberg, R., Ben-Ami, E., Schachter, J. and Besser, M.J. 2013. Nicotinamide inhibits vasculogenic mimicry, an alternative vascularization pathway observed in highly aggressive melanoma. Journal of *PloS one*, *8*(2) : 57160.

[10]. Wardell, S.M., de Souza, M.V., Wardell, J.L., Low, J.N. and Glidewell, C. 2007. Three isomeric 1-(2-chloronicotinoyl)-2-(nitrophenyl) hydrazines, including three polymorphs of 1-(2-chloronicotinoyl)-2-(2-nitrophenyl) hydrazine: hydrogen-bonded supramolecular structures in two and three dimensions. *Acta Crystallographica Section B. Journal of Structural Science*, *63*(1) :101-110.

[11]. Khalil, N.A., Ahmed, E.M., Mohamed, K.O. and Zaitone, S.A.B. 2013. Synthesis of new nicotinic acid derivatives and their evaluation as analgesic and anti-inflammatory agents. Journal of *Chemical and Pharmaceutical Bulletin*, *61*(9): 933-940.

[12]. Hu, R.S., Wang, J., Li, H., Ni, H., Chen, Y.F., Zhang, Y.W., Xiang, S.P. and Li, H.H. 2015. Simultaneous extraction of nicotine and solanesol from waste tobacco materials by the column chromatographic extraction method and their separation and purification. Journal *Separation and Purification Technology*, *146*:1-7.

[13]. T.J.Bandosz. 2015. Catalytic adsorbents obtained from municipal sludges, industrial sludges, compost and tobacco waste and process for their production. United States Patent 13/467027.