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# INCLUSIVE SCENARIO OF NATURAL PRODUCTS OBTAINED FROM AGAR PLANT (AQUILARIA SP.) IN BANGLADESH- A REVIEW

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## **Abstract**

Agarwood is the resinous heartwood obtained from the injured parts of trees under Thymelaeaceae family particularly from *Aquilaria* species. Agarwood is considered as the most prized non-timber forest product (NTFP) used in attars as well as medicines. Quality of agarwood is the determiner for defining its commercial value. Different countries use different grading system to explain the quality of agarwood. In Bangladesh, more than 45 compounds have been identified so far, mostly sesquiterpenoids, alkanes, fatty acid and other volatile aromatic compounds that are responsible for its fragrance and high prices. Ether extract, total phenolic contents, flavonoid, antioxidant and microbiological tests have shown tremendous positive results. Chemical profiling of agar products is the best identified using GC-MS technique by the various authors. Present review discusses the full scenario of *Aquilaria* production, inoculation, extraction methods, determination of agarwood quality and chemical constituents of agar oils and possibilities and barriers of this industry in Bangladesh.

Key words: Agarwood, Agar oil, Cultivation, Microbiology, Phytochemistry

# Introduction

The genus Aquilaria (Lam.) belongs to the Thymeleaceae family that has high commercial, economical and medicinal significance. Aquilaria species are mostly well known for the production of agarwood, a plant natural product extracted from agar tree that is highly valuable and priced product (Chowdhury et al. 2017). Near about twenty-one species of Aquilaria trees are introduced, out of which eight are recognized as agarwood producer (Zich and Compton 2002). These Aquilaria have been cultivated from 3000 years ago in the Middle East, China and Japan (Le 2003) and that is thought native to the Indo-Malaysia region (Mohamed and Lee 2016). Earlier Indian sub-continent was thought as the main source of agar tree for many times but now this tree is cultivated throughout the world especially in Bangladesh, India, Myanmar, Bhutan, Papua New Guinea, Thailand, Laos, Vietnam and so on (CITES 2004). Economically the Aquilaria is important for the agarwood and Agar oil in the worldwide perfumery markets. The most prized part is the aromatic resinous dark colored heartwood of agar tree that is the consequential substance of plant defense counter to external attacks and impacts such as mechanical wound inserted by human being or outer factors, pathological agents, insects and bacteria (Dinh 2010). In Bangladesh, agar (Aquilaria malaccensis, A. sinensis, A. agallocha) is mostly cultivated in the greater Sylhet region and some parts of Chittagong and Chittagong hill tracts (Mohamed and Lee 2016) that has rehabilitated the lifestyles of many people related to

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the sector (Siddiquee 2011). Agar farming is a private sector where more than 30,000 peoples are earning their livelihood under working in 121 agar processing factories that has established in different regions of Sylhet, Bangladesh and has changed their socio-economic status (Abdin 2014). As a profitable form of farming, agar industries are export oriented, highly laborious and being cultivated on commercial basis (Abdin 2014). In Bangladesh, the contribution of these industries to GDP is about \$ 3750000 per year (Siddiquee 2011) that is contributing in poverty reduction. Bangladesh exports their products both as wood chips and oils form. Agar oil is a concentrated volatile aromatic compound extracted from the stem of agarwood plant. Agarwoods are two categories; firstly screw injected agarwood that is common in Bangladesh (Akter et al. 2013) and secondly, naturally insect infested agarwood where insect borer named Zeuzera conferta Wlaker naturally infest agar plant (Aquilaria sp.) (Mazid et al. 2011, Kalita 2015). Literature reports that international market price for agar wood chips ranges from US \$ 20-6000 per kg based on its quality (Akter et al. 2013). Quality depends on so many factors such as color, odor, long lasting aroma and chemical profiles of the oil. To ensure accurate results, chemical analysis of agar oils is used globally for confirming grading of the agarwood (Norazah et al. 2009, Nurlaila et al. 2013). This is massively demanded in several countries which is further treated as perfumery ingredients, aroma enhancers (Norazah et al. 2016), incenses and decorative displays and used as a raw material in traditional and modern medicines (Mohamed and Lee 2016) and so many other different uses. Several researches were done to evaluate the quality of different categories of agarwood sold by the local exporters of Bangladesh. Therefore, the objective of this review is to reveal the overall status of agar plant cultivation and related researches in Bangladesh.

#### **Plantation of Agar tree**

This is an evergreen tree of 49-131 ft. height and 1.0-8.0 ft. diameter (Chakrabarty et al. 1994, Sumadiwangsa 1997) and soft, light, elastic and white to yellowish-white colored wood. It has up to 3.546 inch long oblong-lanceolate leaves with undulating margin. Agar trees grow very fast, and start producing flowers and seeds as early as four years old (Akter et al. 2013). April-May is the flowering period and seeds mature in about three months (Selvan et al. 2014).

## History of agar plant in Bangladesh

Agarwood production was started about 400 years ago at the Suzanagar union under Baralekha upazila of Moulvibazar district in Bangladesh. Earlier agarwood was produced in forest only (Abdin 2014). Due to having inadequate access of agarwood cultivation in forest land, people started social farming system of agarwood in their household lands. Consequently, it became a time consuming process to find out the best quality agarwood. Therefore to mitigate this problem, they developed production of agar oil from ten to fifteen years young trees. Although having such a prolonged history; agarwood sector of Bangladesh does not prosper accordingly due to few policy anomalies and other constraints. Now more than hundreds agar processing factories have already been recognized in different regions of Sylhet, Bangladesh (Abdin 2014) that is going to be a profitable form of farming to contribute in poverty reduction.

#### Present status of agar plant in Bangladesh

Recently agar plant is cultivated on a commercial basis in Beanibazar, Golapganj, Fenchuganj, Sadar of Sylhet, Komolganj, Rajnagar, Srimangal and Barlekha of Moulvibazar district. The Barlekha upazila is one of the major places for growing agar and the Sujanagar union is well known at home and abroad for it. According to the reports of the daily star (2011), about 1,000 families in the village of Sujanagar are earning their livelihood by working in the agar factories set up in their homes and around. Some other mentionable places are also involved in the same work including Saldigha, Bartal, Uttar Sujanagar, Dakshin Sujanagar,

Tangortoli and Rafinagar. These industries have a great contribution to GDP of Bangladesh (Siddiquee 2011) and approximately 25,000 - 30,000 workers are directly involved in agar farming, processing and marketing of agar-based products in the country (Baksha et al. 2009). Agar processing factories have already been established in different regions of Sylhet, Bangladesh (Abdin 2014). Agarwood plantation has been started by BRAC from July, 2007 in Bangladesh at Kaiyachara (17 acre area with 83,400 seedlings), about 700,000 agar seedlings were planted in two nurseries, of 'Kaiya' and 'Sirgasia', at Kaiyachara tea estate because of its economic benefit and conservation value (Akter and Neelim 2008). Additionally, during the period of 1999-2011 Government agar gardens in Denuded forest areas of Sylhet, Chittagong and Chittagong Hill Tracks (CHTs) have been established by the Forest Department (Novel 2017) and Bangladesh Forest Research Institution (BFRI) marginally started same at Charaljani and Keochia research stations in Chittagong (Rahman et al. 2015). Moreover, several small scale and personal agarwood plantations were done at Modhupur (Mymensingh), Birisiri, Haluaghat (Netrokona), Sylhet, Habiganj and Chittagong Hill Tracks (Alam 2004, Islam 2013, Selina et al. 2013). The establishment of this plantation would be significant in terms of the ecofriendly conservation of the endangered species as well as making carbon sink to lessen greenhouse gases. Likewise, there are possible opportunities that would arise in the future for enlightening the livelihoods of meager people in the area by providing income generating means (Selina et al. 2013).

## Agar cultivation in Bangladesh

For sustainable economic development, proper investigation of the current status of agar (*Aquilaria* sp.) cultivation in Bangladesh is indispensable (Talucder et al. 2016). The major agar growing area is south and south-east Asia including Bangladesh, Bhutan, India, Indonesia, Iran, Malaysia, Myanmar, the Philippines, Singapore and Thailand (Oldfield et al. 1998). About eight *Aquilaria* sp. among all fifteen species of the genus are acquainted with agarwood production (Ng et al. 1997) including *A. malaccensis*, *A. agallocha* and *A. secundaria* which are currently the main species for agar cultivation (Broad 1995). In Bangladesh, frequently cultivated species are *Aquilaria malaccensis* (Alam 2004, Chowdhury et al. 2016) and *Aquilaria agallocha* (Bhuiyan et al. 2009, Rahman et al. 2015).

## Climatic and edaphic factors for agar cultivation

Aquilaria species typically grow between altitudes of 0-850 m in locations with average daily temperatures of 20-22°C (Keller and Sidiyasa 1994, Afifi 1995, Wiriadinata 1995). It prefers a subtropical climate with high humidity and rainfall range of 1800-3500 mm (Selvan et al. 2014). Naturally wide range of soils under wide climatic condition including those that are rocky, sandy or calcareous, well-drained slopes and ridges and land near swamps are used for growing Aquilaria sp. (Talucder et al. 2016). Suitable climatic and edaphic factors for agar cultivation were reported in previous study (Talucder et al. 2016) (Table 1). Although it is found in natural forests at an altitude of a few meters above sea level to about 1000 meters, it grows best around 500 meters (Selvan et al. 2014). Judicious quantity of sunshine and well-drained soil are the necessities for most of the agarwood saplings although they can grow under some shady condition in plantation (Selvan et al. 2014, Rima et al. 2015). Actually, sloppy lands are endorsed for agar plantation (Jansen 2003) as significant mortality rate was reported due to water logging (Akter and Neelim 2008). In Northern-East part of the country Aquilaria have been successfully implanted in plantations (Blanchette et al. 2015). Around 75% of the plantations have been established on degraded land as well as denuded hills and rest 25% were established at the homesteads of farmers (Rahman et al. 2015). Agar tree can also be grown

on border side of field, garden, roads, school and office compounds; along bankside of ponds, tanks, canals; parks and residential sites. In hilly areas/tillas as in Barak valley it can be planted on poor soils on hill slopes, tilla tops.

**Table 1.** Suitable climatic and edaphic factors for *Aquilaria* species cultivation

Climatic variables and soil factors	Range of values
Altitude range	29 - 1000 m
Annual rainfall	1500 - 6500 mm
Annual temperature	22 - 28°C
Maximum temperature of hottest month	22 - 40°C
Minimum temperature of coldest month	14 - 22°C
Absolute minimum temperature	5°C
Soil texture	Light; medium
Soil drainage	Free
Soil reaction	Acid; neutral
Special soil tolerances	Shallow; other

Source: Adopted from Talucder et al. (2016), Akter and Neelim (2008).

## Propagating material and plantation technique

Talucder et al. (2016) reported that there was no other modern technique of propagation except conventional propagation by seed in Bangladesh. Multiplication of the plant occurs through mature seed (Ahmed 2010). Seeds are recalcitrant, have low period of viability (7-10 days) which becomes available in the month of June-July (Ahmed 2010, Talucder et al. 2016). Immediately after the fruit opening, seeds are collected for plantation because of difficulty in seed storage due to quick loss of viability. Sand beds are used for seed germination following transferring to polybags (Anonymous 2004, Talucder et al. 2016). Seed germination process is epigeal indicating necessity of special nursery management care. After just 25 days of cotyledons drop down, seedlings are transplanted into poly bags organized under temporary shade (Talucder et al. 2016). Poly bags are then arranged in bamboo pole surrounded beds with adaptation of the normal management practices. To avoid the penetration of roots into the soil, bags are shifted at monthly interval followed by light watering. Agar trees were planted in monoculture and block plantations (Rahman et al. 2015). After cleaning the planting sites, standard-size pits were dug at a spacing of 2 m × 2 m and each pit was left unrestricted for at least 15 days after placing 1.5-2.0 kg of cowdung. Inorganic fertilizers can also be used during establishment of plantation. One-year-old seedling usually planted in April or May. Rahman et al. (2015) found that the conventional management practices were adapted mostly based on local knowledge and technology to manage agar plantations. Weeding to reduce the competition between the agar trees and other undesired plant species was the most frequently applied operation in agar plantations. Intensive weeding was carried out in the first 3 years. Staking was not a common practice in agar plantations (Rahman et al. 2015). Mulching at the bottom of agar plant is the most frequent practices for water loss reduction. During the seedling and sapling stages both organic and inorganic fertilizers were applied to promote growth. The organic fertilizers included fresh cowdung, plant debris and compost made from cowdung. The common inorganic fertilizers used in agar plantations included urea, tripple superphosphate (TSP) and muriate of

potash (MP). In order to protect agar trees from leaf sucker insects, farmers used pesticides including malathion and endrin. Pruning was done only once a year, from the third to fifth or sixth year after out planting. According to the farmers, the best time for the pruning is the end of May. Thinning was rarely practiced in agar plantations. Farmers used boundary fencing to protect seedlings from herbivores and other disturbances (Rahman et al. 2015). About 12-15 years are required for agar wood tree to be used as raw materials. Ironing (putting iron rod inside the tree, about 100-150 kg iron rod/medium sized tree) is done at 8-10 years old tree. Then after 3-4 years, agar tree become suitable for agar oil extraction (Abdin 2014).

# Agarwood

Agarwood sometimes known as eaglewood is the resinous heartwood of agar tree, relatively light and pale colored, formed in response to natural infection (such as pathological infection by the insects, bacteria etc.) or artificial induction like, mechanical injury (Akter et al. 2013). The wood is very condensed and dark when it is being matured and produced a dark aromatic resin (Blanchette 2006, Chowdhury et al. 2017).

## Inoculation methods of agar plants in Bangladesh

The healthy woods or white wood of *Aquilaria* trees are without oleoresins. Agarwood farmers from different Asian countries are tried several wounding methods to produce agarwood, including chopping, nailing, holing and trunk breaking (Ahmad et al. 2017). In Bangladesh, generally two categories of agarwood (*Aquilaria* sp.) are produced by the agar workers. One is artificially screw injected agarwood (Akter et al. 2013) and the other is naturally insect infested agarwood (Hoque et al. 2019). Broadly, inoculations of agar trees are of two types- i) natural inoculation and ii) artificial inoculation.

#### **Natural inoculation**

Under natural conditions, oleoresin can only be produced by natural wounding such as injury by external factors, insect attack or bacterial/fungal infection around the wounded or rotting parts of the trunk (Blanchette 2006). Naturally agarwood production quantity is low (Gibson 1977). Naturally agar plant (*Aquilaria* sp.) is infested by insect borer *Zeuzera conferta* Wlaker (Kalita 2015, Hoque et al. 2019) in the wood when trunks of standing trees are bored by the larvae (Fig. 1a) (Selvan et al. 2014). It is seen that insect larvae bore the standing tree trunk of *Aquilaria* and make tunnels inside the tree trunks (Fig. 1b) (Hoque et al. 2019). Fungus (such as *Fusarium* sp., *Rhizopus* sp., *Aspergillus* sp., *Mucor* sp. and *Penicillium* sp.) can enter the plant through the erect hollow/zigzag channel inside the stem, which work for easy extent of infections and responsible for physiological host-parasite interaction (Selvan et al. 2014). After time being, infection can spread on all sides slowly and progressively and finally a large wood volume gets infected. An addition of oleoresins goes on increasing with the increase of infection rate as well as matures of infection. The more of oleoresins are deposited the concentration of color of the infected wood increases and to end with becomes black and odoriferous due to upsurge in concentration (Selvan et al. 2014).





Figs. 1(a-b): Natural inoculation of agarwood. (a) Zeuzera confetra larva, and (b) Naturally insect infested agarwood.

#### **Artificial inoculation**

Artificial inoculation technique has found to be most productive and reliable method for the enrichment of agar production (Hoque et al. 2019). In Bangladesh people are practicing screw injected method so that they can produce resin artificially (Rahman et al. 2015). In this method, agarwood tree is wounded by hitting of iron nails (Rahman et al. 2015) into the trunks spirally with hundreds to thousands of nails put into each tree (Fig. 2a) (Blanchette et al. 2015). Screw is metal that is injected into the trunk of agar plants. Plant secrets resin in uniform pattern to protect themselves from external attack and contain different chemicals known as secondary metabolites (Fig. 2b) (Mazid et al. 2011, Hoque et al. 2019). Metallic component is unable to synthesis any chemical during its injection in agar tree. After many years, each metallic wound produces a minor amount of low quality resinous wood (Hoque et al. 2019). Moreover, agarwood produced from this practice is generally of inferior quality and unable to meet the preferred market demand (Persoon 2007). In the situation of increasing demand and price, some efforts are needed to stimulate agarwood production artificially as well as to drive a quicker process of its formation (Ahmad et al. 2017).





**Figs. 2(a-b):** Artificial inoculation technique of agarwood. (a) Screw injected into agar plant, and (b) Resin produced in screw injected agarwood.

# Agar oil extraction method

As agar products are traded both as chips and oil form, extraction of agar oils from agarwood has a great importance. Generally most of the trade essential oils are volatile, practically stable to the action of heat and insoluble in water therefore are fit for processing by distillation. There are three methods (Baksha et al. 2009) through which agarwood oils can be extracted namely, 1. Hydro-distillation, 2. Steam distillation and 3. Super critical CO<sub>2</sub> extraction. Out of three, the method practices in Bangladesh are Hydro-distillation method (Fig. 3) to extract agarwood essential oil (Hoque et al. 2018) which is the simplest and cheapest process of distillation (Chen et al. 2013). Essential oils are extracted from healthy or artificially injured plants using this method. At first, wood containing resin were chopped into wood chips and then soaked into water for 10-12 days (Hossen and Hossain 2016).



Fig. 3: Hydro-distillation chamber of agar oil extraction.

After this, the soaked chips were taken out, shattered and pounded manually or in machine. The chips were put in mini plants or Retort (Deg), where it is boiled constantly for 4-5 days. The heat and steam cause the cell structure of the plant material to burst and break down, thus freeing the essential oils (Chen et al. 2013). After oil vaporization, condensation occurred in distillation pipe (condenser) (Xiaojin et al. 2017). The extracted liquid is a mixture of oil and water is settled on water in collection pot (Hossen and Hossain 2016). Essential oils are easily separable from water and siphoned off for being water insoluble. They have the capability of floating on the surface because of being lighter compared to water. Then the sample was collected manually, sundried and bottled. Best quality comes from 1st distillation. A flow chart showing agar oil extraction process is given at Fig. 4 (Chen et al. 2013, Hossen and Hossain 2016, Hoque et al. 2018).

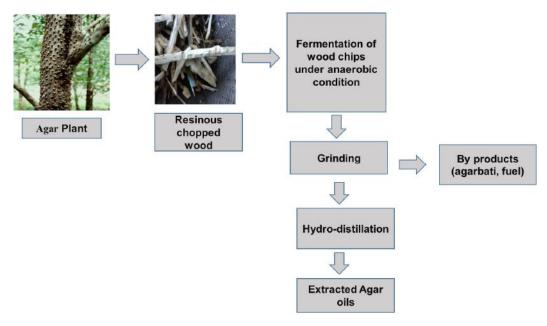


Fig. 4: Flow chart of agar oil extraction process.

#### Agar oil

Agar oil is highly intense volatile fragrant compound extracted from the heartwood of *Aquilaria* plant. Agar oil is used as incense, perfumery and for traditional medicines (Pripdeevech et al. 2011). It is an important ingredient in the perfumery industry and to generate aroma in wedding ceremonies or banquets (Yusoff et al. 2006, Takemoto et al. 2008, Norazah et al. 2012, Zhang et al. 2012). Multiple usages have been found from the agarwood trunk, branches, chips and flakes of uniform quality powder to essential oil (Wetwitayaklung et al. 2009). Many studies have shown that there has been a dramatic increase in agarwood oil usage in Malaysia (Najib et al. 2011, Tajuddin et al. 2013, Desa et al. 2021). Similarly there is an increasing trend of agarwood plantation in this country and also the market demand of its oil is enhanced (Hamid 2011, Rashid and Zuhaidi 2011).

#### Chemical analysis of agar products of Bangladesh

# Ether extract, total phenolic, flavonoid and antioxidant status

Agarwood samples produced high percentages of ether extract which indicates its oil producing capabilities. Results from Bangladeshi wood samples of insect infested and nail injected woods showed fluctuations may be due to different level of insect infestation and using higher mass of insect infested wood sample (Ahmed et al. 2019) (Table 2). More insect injuries can produce higher oils (Hare 2011), phenolic, flavonoid contents and secondary metabolites which may contribute to the long oil shelf-life, color and several organoleptic characteristics (Bulotta et al. 2014, Hoque et al. 2019). Insect create initial site of infection of microbes such as-fungi and gradually moves upward (Kalita 2015, Hoque et al. 2019) that might have positive correlation between beneficial insect attack with phenol and flavonoid production (Zhao et al. 2018, Hoque et al. 2019). Microbes can also grow around the nail that can helps to produce black resinous oils (Zhang et al. 2010,

Kalita 2015) but the percentage of natural microbes attack in nail injected agar tree is very low (Kalita 2015, Hoque et al. 2019) as metal (nails) is unable to secret any chemical during its injection in agar tree.

Table 2. Ether extract, total phenolic, flavonoid and antioxidant status of agar wood/oil in Bangladesh

Analysis types	Insect infested wood	Screw/Nail injected wood	White wood	Reference (s)
Ether extract (%)	11.08 ± 0.94	20.49 ± 0.04	1.80 ± 0.03	Hoque et al. (2019)
	$18.90 \pm 0.60$	11.03 ± 0.19	$1.84 \pm 0.04$	Ahmed et al. (2019)
Phenolics (mg/g)	2.97 ± 0.07	$3.6 \pm 0.052$	$2.55 \pm 0.05$	Hoque et al. (2019)
	$3.5 \pm 0.06$	$2.98 \pm 0.07$	$2.50 \pm 0.05$	Ahmed et al. (2019)
Flavonoid content in μg QE/ml	$7.82 \pm 0.23$	$6.58 \pm 0.62$	-	Ahmed et al. (2019)
Antioxidant activity IC50 (µg/ml) of agar oil	-	0.904	-	Ahmed et al. (2019)

## Phytochemistry of agar (Aquilaria sp.) in Bangladesh

Chemical constituents of the infested agarwood oils are different from the artificially nailing wood of Aquilaria (Bhuiyan et al. 2009). Present review focuses on the chemical constituents of agar oil from the cultivated agar wood in Bangladesh. More than 45 compounds have been identified (Table 3) so far, mostly sesquiterpenoids, alkanes, aromandendrene and other volatile aromatic compounds (Bhuiyan et al. 2009, Hoque et al. 2018). Researches on exportable agar oil of Bangladesh revealed that sesquiterpenes and aromatic compounds occupied the major portion (above 90 %) and the rest portion is occupied with minor alcoholic and fatty acid group (Bhuiyan et al. 2009, Hoque et al. 2018). Sesquiterpenes is 15-carbon skeletons further classified into selinane, nootkatane, cadinanes, guaianes, prezizanes and agaropsiranes categories based on the basic structure. Sesquiterpenes are thought to be less volatile than other terpenes, have a greater potential for stereo-chemical diversity and have stronger odors. They are anti-inflammatory (Jeena et al. 2013), strong antioxidant (Ornano et al. 2013), anti-tumor activity (Park et al. 2011) and have bactericidal properties (Ishnava et al. 2013). Sesquiterpenes in the agarwood of Bangladesh is 94-95% which is found higher than Chinese 80-89% (Chen et al. 2011), Malayasian 80-85% (Nor et al. 2015) agar oil (Bhuiyan et al. 2009, Hoque et al. 2018). Alcoholic compound such as 3-Tetradecyn-1-ol and 2-Octylcyclopropene-1-heptanol possess bactericidal rather than bacteriostatic activity against vegetative cells (Raja et al. 2011). Occurrence of 4-phenyl-2-butanone compound, responsible for waxy herbal odor type medium odor strength (Yumi et al. 2014).

Table 3. Chemical compounds identified in agarwood

SI. No.	Chemical constituents	Chemical structure	Reference(s)
1	4-phenyl-2-butanone	O CH <sub>3</sub>	Hoque et al. (2018)
2	N-benzyloxycarbonyl-serine, methyl ester	HNIIITE OH	Hoque et al. (2018)
3	6,7-dimethyl-1,2,3,5,8,8a- Hexahydronaphthalene		Hoque et al. 2018
4	Ledene oxide- II		Hoque et al. (2018)
5	Aromandendrene	H H	Bhuiyan et al. (2009), Hoque et al. (2018)
6	2H-3,9a-Mehtano-1-benzoxepin,octahydro- 2,2,5a,9-tetramethyl		Hoque et al. (2018)

7 Tricyclo [5.2.2.0(1,6) undecen-3-ol,2-methylene-6,8,8-trimethyl

Bhuiyan et al. (2009), Hoque et al. (2018)

8 Tricyclo[4.4.0.0(2,7)]dec-8-ene-3-methanol,  $\alpha,\alpha,6,8$ -tetramethyl-

Hoque et al. (2018)

9 4-Isopropenyl-4,7-dimethyl-1-oxaspiro[2.5]octane

Hoque et al. (2018)

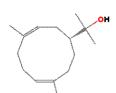
Bicyclo[4.4.0]dec-5-ene,1,5-dimethyl-3-hydroxy-8-(1-methylene-)

Hoque et al. (2018)

11 1,2,3,4,5,6,7,8-Octahydro- $\alpha$ , $\alpha$ ,3,8-tetramethyl-5-azulenemethanol

Bhuiyan et al. (2009), Hoque et al. (2018)

12 3,7-Cyclodecadiene-1-methanol,  $\alpha$ , $\alpha$ ,4,8-tetramethyl-, [s-(Z,Z)]



Hoque et al. (2018)

13 Cyclohexanemethanol,4-ethenyl- $\alpha$ , $\alpha$ ,4-trimethyl-3-(1-methylethenyl)-, [1R-(1 $\alpha$ ,3 $\alpha$ ,4 $\beta$ )]-

Hoque et al. (2018)

14 Isoaromadendrene epoxide



Hoque et al. (2018)

15 Tetradecyn-1-ol



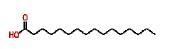
Hoque et al. (2018)

16 2-Octylcyclopropene-1-heptanol



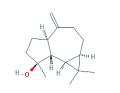
Hoque et al. (2018)

17 n-Hexadecanoic acid



Bhuiyan et al. (2009), Hoque et al. (2018)

18 (-)-Spathulenol



Bhuiyan et al. (2009)

19 6,9-Octadecadiynoic acid, methyl ester

Bhuiyan et al. (2009)

20 2-Naphthalenemethanol, 1,2,3,4,4a,5,6,7-octahydro-.alpha..alpha.4a,8-tetramethyl-, (2Rcis)-

Bhuiyan et al. (2009)

21 β-Patchoulene

Bhuiyan et al. (2009)

22 Naphthalene, 1,2,3,5,6,7,8,8a-octahydro-1,8a-dimethyl-7-(1-methylethenyl)-, [1R- $(1\alpha,7\beta,8a\alpha)$ ]-

Bhuiyan et al. (2009)

23 Azulene,1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-,[1R- $(1\alpha,3a\beta,4\alpha,7\beta)$ ]-

Bhuiyan et al. (2009)

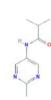
24 Aristolene

Bhuiyan et al. (2009)

25 Eremophilene

Bhuiyan et al. (2009)

26 5-Isobutyramido-2-methyl pyrimidine



Bhuiyan et al. (2009)

27	Isolongifolene, 9,10-dehydro-		Bhuiyan et al. (2009)
28	Diphenoxylic acid	H O N N N N N N N N N N N N N N N N N N	Bhuiyan et al. (2009)
29	Neoisolongifolene, 8,9-dehydro-		Bhuiyan et al. (2009)
30	Isolongifolen-5-one		Bhuiyan et al. (2009)
31	Naphthen-1-acetic acid, 8-methoxyalpha methyl	N. O	Bhuiyan et al. (2009)
32	3-Hydroxy-7-methoxy-2-naphthoic acid	CH₃O OH	Bhuiyan et al. (2009)
33	Propanoic acid, 3-(diisopropylphosphino)-, methyl ester		Bhuiyan et al. (2009)

34 3-Methoxy-6,7,8,9-tetrahydro-dibenzofuran-2-ol

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Bhuiyan et al. (2009)

35 Cadinene

Bhuiyan et al. (2009)

36 Longiverbenone

Bhuiyan et al. (2009)

37 Caryophyllene oxide

Bhuiyan et al. (2009)

38 (6-Hydroxymethyl-2,3-dimethylphenyl)methanol

Bhuiyan et al. (2009)

39 9-[4-[1,3-Diphenyl-2-imidazolidinyl]-2,3-O-[1-methylethylidene]-.beta.-d

Bhuiyan et al. (2009)

40 α-Cedrene oxide



Bhuiyan et al. (2009)

41	Viridiflorol	H	Bhuiyan et al. (2009)
42	Octacosane		Bhuiyan et al. (2009)
43	Diisooctyl phthalate		Bhuiyan et al. (2009)
44	7-Isopropenyl-4a-methyl-1- methylenedecahydronaphthalene		Bhuiyan et al. (2009)
45	Cycloheptane, 4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1-vinyl		Bhuiyan et al. (2009)
46	Caryophyllene oxide	$H_3C$ $H_3C$ $H_3C$ $H_3C$ $H_3C$ $H_3C$ $H_3C$ $H_3C$	Bhuiyan et al. (2009)

# Microbiological test of agar products

Recent microbiological test of agar product was conducted by the researcher on antimicrobial activity test of the exportable agar oils from Bangladesh against some specific bacterial strains such as- *Escherichia coli, Staphylococcus aureus, Salmonella* and *Vibrio* (Hoque et al. 2018) and a bacterial isolate was identified for enhancing fermentation process of wood (Ahmed et al. 2019). Antimicrobial sensitivity test was done using agar well diffusion method and the results indicated non-significant sensitivity against all the tested bacteria

(Hoque et al. 2018). On other hand, among tested bacteria - *Bacillus* sp., *Klebsiella* sp., *Aeromonas* sp., *E. coli* and *S. aureus*, only *Bacillus* sp. may contribute to increase the fermentation of agarwood to surge agarwood yield (Bhore et al. 2013). Chen et al. (2011) reported that if the active components from agar oil can be isolated and purified, their antimicrobial activities could be stronger.

## Agar quality analysis technique

There is no internationally common system for ranking agar wood each sourcing country may have their own grading system or none at all (Hoque et al. 2018). These grades may not be accurate or inaccurate, but must be acceptable at both ends of the deal (Rozi and Shiou 2016). In Bangladesh, generally people categorized agarwood products according to its color, fragrance and fixative based on human's sense. Although this is a complex way to standardize its quality from the scent due to human nose cannot smell many tasters continuously (Norazah et al. 2009, Rahman 2009, Pripdeevech et al. 2011). There are several techniques, firstly, color is an indicator of oil or wood quality (Jayachandra et al. 2014) for example, dark color refers to the high quality because of high resin deposition (Boon et al. 2016) with very expensive market price (Norazah et al. 2008). Secondly, "sinking test" whereby wood chips have to drop in water to relate with the wood's quality. Wood that floats are considered as inferior in quality due to the low resin content (Rozi and Shiou 2016). Thirdly, "burning test" in which buyers can assess the aroma of agarwood by burning a small sample of the agarwood chip (Rozi and Shiou 2016). Fourthly, the long-lasting fragrance indicates that the quality is superior and naturally it influences the grade and pricing (Antonopoulou et al. 2010). Fifthly, the geographical source or place of origin and the characteristics of the agarwood including the size, shape, age and parts of the tree from where it is derived (Compton and Ishihara 2004, Wyn and Anak 2010). Sixthly, taste such as sweet, sour, salty, hot, and bitter, which turns grading into a very subjective matter and people dependent (Rozi and Shiou 2016). Therefore, different ways are available for grading or classifying the agarwood oil used in the country. Hence, no standard is available for high and low quality determination (Tajuddin 2011). Beside this grading system there are some other grading systems which are used by the local peoples dealing with trading and sell of agarwood. The most reliable and acceptable technique for the confirmation of agar oil is the chemical profile analysis (Hoque et al. 2018). Different color has different chemical constituents (Tajuddin and Yusoff 2010, Pornpunyapat et al. 2011). For an example, GC-MS analysis found that oxidoagaro chromone as a pale yellow in oil component (Yagura et al. 2005). Both color and odor have a complex mixture of sesquiterpenes and chromone derivatives (Tajuddin 2011). As for being major volatile components, the presence of sesquiterpenes is popular in market (Wetwitayaklung et al. 2009). The exportable oils from Bangladesh contained a complex mixture of different secondary metabolites (Hoque et al. 2018). Researches revealed that sesquiterpenes content used as grade indicator can be obtained only by chemical analysis of agar oils (Norazah et al. 2009, Nurlaila et al. 2013).

#### Uses of agar products

Agar products have many uses in industry as well as social purposes that have high economic values (Table 4).

**Table 4.** Table showing uses of agar products

Agar products	Uses	Reference(s)
Agarwood chips	Raw materials in fragrance industry	Abdin (2014)
Agar oil	Body Fragrance, clothes fragrance and raw materials in the perfume industry	Antonopoulou et al. (2010), Abdin (2014), Norazah et al. (2009)
Agarwood chips, oudh, Bakhoor, Scented chips	To receive honored guests	Antonopoulou et al. (2010)
Agarwood raw powder	Incense production and burning powder during prayers in Buddhist temples	Abdin (2014)
Agarwood beads	Religious purpose	Abdin (2014)
Agarbati	Scented flame in religious purpose	Hossen and Hossain (2016)
Unused branches	Firewood	Hossen and Hossain (2016)
Agarwood medicines	Several types of crude medicine, anti- inflammatory, unanai ayuravedic, nervous disorders, stimulants, stomach tumors	Abdin (2014), Mohamed and Lee (2016), Abas et al. (2017), Yadav et al. (2013)
Liquor with agarwood	Liquor products produced by the Taiwan Tobacco and Liquor Corporation	Song (2002)

# Agar industry in Bangladesh: possibility and barriers

Present literatures have revealed very little amount of knowledge about agar industry in Bangladesh. Merely a few studies were being carried out like Akter and Neelim (2008) and Abdin (2014). It is totally an export oriented industry depending on the local raw materials and indigenous technology. In Bangladesh, more than 100 enterprises are involved in the production of agar wood and agar-oil which are mostly centered at Baralekha upazila of Moulvibazar district. Although having no availability of official export data, it is estimated that from Bangladesh local entrepreneurs are exporting about Tk. 5.00 million to Tk. 100 million (the stakeholders consultation meeting) per year from agar wood or agar-oil SME Foundation 2013 (Abdin 2014). Depending on topographical location and cultural deposition, first grade agar wood selling as woodchips, wood pieces, powder, dust, oil, incense ingredients and perfume is very costly (La Frankie 1994, Barden et al. 2000, Gunn et al. 2004, Compton 2007). Due to lack of diversified knowledge, Bangladeshi agar sector is producing only three items namely; agar wood chips, agar wood oil and agar wood powder/dust (Abdin 2014). Pricing of agarwood chips start at \$30-\$9,000 per kg based on resin content inside the chips (Babatunde 2015). Agar oil is also highly priced selling at US \$ 30,000 per kg (Chowdury et al 2016). The present global market is projected to US \$ 6 to \$ 8 billion for agar oil and other related agar products (Akter et al. 2013) and the commercial purchasers of agar oil is expected to exceed US \$ 36 billion in 2017 (http://www.ouddh.com = 2065877). In spite of the scenario, agar sector still remains as an unofficial/informal sector in Bangladesh. Lacks of governmental supports as well as limited governmental ingenuity to formalize the sector are the major constraints for industrializing agar products in Bangladesh. Currently, private sectors use their raw materials (trees) from their own gardens although governmental and social forests were the

major primary source formerly. Also high import duty charge by the importing countries, limited access to the government forests while selling trees by the local agarwood entrepreneurs and lack of training facilities are also paving the way for agar industrializations (Abdin 2014, Chowdhury et al 2016, Das et al. 2018). Scarcity of high quality seedlings (Rahman et al 2015, Das et al 2018) and labor scarcity (Das et al. 2018) also have some contribution to make this sector unproductive. If this sector is prioritized as a promising one by the government of Bangladesh, relevant agencies and required steps are taken to mitigate the obstacles, then obviously the economy of Bangladesh will experience a huge flourishment.

#### Conclusion

Agar trees have attracted attention for its economic prices and scarcity. Agar is the most luxurious valued items of the world that could be one of the main sources for earning foreign currency and enrich export figure of Bangladesh if the country can flourish this sector. Data on exportable essential oils from Bangladesh is vital for receiving required market price and grade of the crops in the global market. As naturally infested agarwood is expensive than artificially inoculated agarwood, the production could be a multidimensional field in the prospects of Bangladesh through practicing the both techniques. The hilly fallow area of the country could be shifted to treasure house. Although numerous NGOs and private owned plantations have been financing for *Aquilaria* plantation in a number of districts in Bangladesh but the achievement of the effort basically depends on the agar wood production. Effective artificial induction method is needed to be introduced. This is the key time to think about financing on exploration of induction to reach economic goal of plantation. The selection of new plants from authentic sources, ideal inoculation and induction methods could spark the economy through quality enhancement. A multidisciplinary tactic could be introduced with the expert professionals of forestry, mycology, biochemistry and microbiology to achieve the goal.

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#### Competing interests

Authors have declared that no competing interests exist.

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