

PHARMACOGNOSY AND PHYTOCHEMISTRY

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Week 4

Lecture 20

Distribution of tannins (Part 2)

Hello everyone, and welcome to the NPTEL course on pharmacognosy and phytochemistry. Welcome to the session on tannins. In the previous session, we studied a few examples of tannins. Just to summarize, we studied cases of hydrolyzable tannins, which included nutgalls, amla, pomegranate, myrobalan, both harada and baheda, as well as the Malabar kino.

In this session, we will learn about condensed tannins. Do you remember? Condensed tannins are the category of non-hydrolyzable tannins, and they have more of either a phloroglucinol type or a flavonol type nucleus. In this particular session, we will study pale and black catechu, which are examples of flavonol-type condensed tannins. So, let's start with the pale catechu.

Now pale catechu often referred to as Gambier or pale catechu or catechu is an aqueous extract. Unlike other drugs we have studied so far, which contain the whole plant part referred to as organized drugs, in this case, the pale catechu is an example of an unorganized drug, meaning the drug has no cellular units. What we take from this plant—the tree is called *Acacia*

catechu—is the aqueous extract from young twigs, which is dried to develop a hard, stony material. This material is referred to as pale catechu because of its paler color than the subsequent drug we will be dealing with, which is black catechu. The pale catechu comes from the Malayan region. Now this includes Malay countries such as Indonesia, Malaysia, Sumatra, Borneo, or even Singapore.

These are the regions where the plant occurs very commonly. So what is done is, in order to get this extract, you cultivate the plant. Generally from the seedlings, and in its second year when it has developed sufficiently, young twigs or young shoots, you collect the first crop of it. Now the good part is, after you collect the first crop, the plant will still keep on growing new shoots, so every year you can collect fresh produce,

and it is said that you can collect it for up to 20 years of the plant. And the best produce is thought to be somewhere around eight years. So that's where your tannin content or your extract, which comes from this plant, is maximal. So this plant, the fresh twigs or the young shoots, are cut off. They are collected.

They are put together in a metal drum with about three-fourths of water. So once it is submerged nicely with three-quarters of water, you can start boiling it. Now this is boiled for a period of about three hours. When it is boiled, you will see that gradually, a greenish-brownish color extract comes out. Now since you have this material—this material like your tea powder—often interferes, so large

wooden forks or meshes are taken, and this raw material or the cellulosic material is moved toward the side. Now as you move that material toward the side, you will see that that also takes along some juice or extract with it. Now if you want to increase the efficiency, this material is pressed off. with those same wooden forks so that the juice flows back into the vessel

sometimes this mark is also washed so you can add additional water on this mark and whatever is there the washings are also allowed to flow in the container in such way that the plant material from the plant material you can extract maximum tannins and all these tannins go on in your vessel Now once you have the extracts and all the washings being done in the vessel you could concentrate it.

Now this concentration is done for more than an hour till you get a very pasty syrupy consistency of a liquid. Now this thick yellow viscous liquid or paste is then put in troughs a wooden troughs and this wooden troughs are actually kept in some cold stream of water what happens is as the juice is hot it is still fluid but the moment you put it in a flowing stream of cold water it starts thickening This is allowed to thicken and as it has

you know as it gains a good pasty consistency this is then put into trays or drums or even vessels. So once that is done on complete cooling it will solidify. Once it solidifies it is cut with the help of a blunt wooden knife to get what are called as cubes of catechu. Now this pale catechu is a very good source of tannins. So if you see the chemical composition it contains monomeric that is

pseudotannin, catechin almost 7 to 30 percent depending upon the age, depending upon the geographical location. Changes might happen but on an average it has about 7 to 30 percent of catechin. So much so that if you take it, break the catechu block and mount it in lactophenol you would even see the catechin crystals. Apart from that, it also has polymerized catechin.

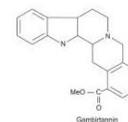
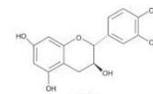
If you remember, condensed tannins are formed when this flavonol unit of catechin starts polymerizing it in itself by forming numerous units. So, these polymeric units are called phlobatannins or catechu tannic acid because the monomeric unit is catechu. So this drug has about, you know, between 25 to 55 percent of phlobatannin. You can call it catechu tannic acid.

Now, it has one more interesting principle, and that is called gambier fluorescent and catechu red. Now, gambier fluorescent, as the name indicates, has a little ability to fluoresce. Catechu red is more of a phlobaphene derivative, and quercetin is a flavonoid derivative. Apart from that, it contains indole alkaloid. You can see the structure of it.



Pale catechu

- 7 to 30% of pseudotannin catechin and
- 25 to 55% of a phlobatannin catechutannic acid.
- It also contains catechu red, gambier fluorescin and quercetin.
- It contains indole alkaloid up to 0.05%, which includes gambirtannin and its derivatives.



So this is the indole moiety. In the indole alkaloid, this is called gambir tannin. Now, gambir tannin is also a principle responsible for the fluorescence of the pale catechu extract. Now, this being a condensed tannin, you can evaluate it using numerous methods. Why evaluate it? Because in the market, pale catechu and black catechu are often admixed with each other, and

that is the reason it is often crucial to identify pale catechu. Now, how do you identify pale catechu? So, if you break it and, as I told you, if you mount it under a microscope with a little lactophenol or a mounting solution like water—simple water—you will see circular crystals of catechin under the microscope.

Apart from that, if you see a compound called gambir tannin or gambir fluorescence, that can also help you identify pale catechu. So, how do you identify it? How do you see this fluorescence? Now, we know this compound is an indole alkaloid, and alkaloids in their basic state. That is, alkaloids in their unionized state have excellent solubility in nonpolar solvents.

So, what do you do here? You take your pale catechu. Now, this pale catechu—basify it a bit. You can basify it with ammonia, sodium hydroxide, or even potassium hydroxide. The intention here is to create a basic pH so my alkaloid remains in a unionized state.

The moment it is unionized, its solubility in water drops, but its solubility in organic solvents is enhanced. And as a result, if you take this catechu, alcoholic extract and partition it using petroleum ether, the petroleum ether layer will happily absorb. That is, your gambiritanin—the alkaloid—will partition into the petroleum ether layer. This petroleum ether layer, if seen under UV light, will show a nice green fluorescence.

The presence of that green fluorescence indicates that the sample you have is pale catechu. Now, another interesting test to identify pale catechu is a test for chlorophyll. In this case, when you saw the raw material, it was the twigs—the fresh leaves and fresh stem. If you see fresh leaves and stems, which are the apical part of our tree,

they are greenish in color, and this green color is attributed to a pigment called chlorophyll. Now, chlorophyll is a non-polar lipophilic pigment. So what you have to do is again take

pale catechu powder, crush it nicely, or take a pale catechu slab and crush it nicely. Once in the powdered state, you extract it with organic solvents such as chloroform. Generally, chloroform is used because chlorophyll dissolves readily in chloroform.

You will see most of the pigments, including your chlorophyll, will dissolve in the chloroform layer. Now filter it so that the cuttage doesn't interfere, and to that filtrate, if you evaporate it in a white or clear dish, you will see a greenish-colored residue. This greenish-yellow residue is due to the presence of chlorophyll and oxidation products of certain carotenoids. So this is also another unique test that will help you identify pale catechu.

The other tests which it will obey are definitely your vanillin hydrochloric acid test for condensed tannins, as well as the matchstick test which we studied previously. Now, applications: where is pale catechu used? Now, pale catechu, owing to its very high content of catechin, is used as a local astringent. So, for mouth ulcers, it is used.

It can be used even topically in creams. It is given to patients suffering from diarrhea as a general astringent. And in some countries, especially the Malayan countries where it commonly grows, it is given with betel leaf. Now a large amount of it initially was used in tanning and dyeing industries. We know your tannins are very good pigments which have a staining ability especially for proteinaceous fibers.

So this has been used again classically for the dyeing and tanning industry again for converting your animal hide into leather. Now, another species that we study is a little Indian species called as black catechu. Black catechu, if you have to say the Indian catechs, you remember katha or katha, which is there in your paan masala. It is the same black catechu I am referring to. So it's often called cutch, black catechu or katha.

It comes from a plant that is your, again this is an unorganized drug. It is coming from the aqueous extract. So the drug here is not a cellular one. The drug here is the aqueous extract which has been obtained from the heartwood of *acacia catechu*. Now this belongs to the family Leguminosae.

As compared to your black catechu where you used your young twigs or young shoots let me point out that in this case it is the heartwood which is used. So if I have to tell you what heartwood is, if you take a tree trunk and dissect it the outer part of it is called as bark so this is the bark. This is what we call it as the sapwood and this is what we call it as the heartwood.

So the heartwood is the oldest part of the wood and it's rich in secondary metabolites. So your black catechu or katha comes in from the heartwood of the *Acacia catechu* tree. Now this is again grown abundantly in the deciduous forest as well as cultivated in Punjab or regions from Punjab to Assam, Madhya Pradesh, Maharashtra, Gujarat, Rajasthan, Bihar and Tamil Nadu. Now this being a hot wood, it's a little difficult to procure.

So once you have a tree which is completely matured, you have to cut the tree. Completely or in some cases coppicing is done and a little stump is left over the ground. The rest of the tree is cut off after that particular height of about say one to three feet. Once this coppicing is done the outer bark is peeled off then the sapwood is peeled off and the inner heartwood is taken. Now this inner heartwood is cut into finer chips.

Why? Because you want to prepare an aqueous extract for it. If you keep the whole wood or log as such, the extraction will not be efficient. So in order to increase the surface area, get more efficient extracts or better rich extracts, you have to chip it and make finer particles of it.

This is generally done nowadays with sawmills where you have good advanced cutters. So this hardwood is taken, cut into fine little chips, and these chips are then taken and boiled in earthen pots. So this is the traditional method. Nowadays, even stainless steel pots have been used, which are nicely jacketed to create a good temperature or heat. Traditionally, these earthen pots were heated with the sapwood itself so that no part of the wood goes to waste. Now, this

process is continued until you get a syrupy mixture again. The good part is that the chips can easily be separated, so it is strained through a cloth and then poured into an iron pot with continuous stirring. Now, why is this iron pot specifically put in here? If you see catechu, especially black catechu, it is rich in condensed tannins.

But remember that condensed tannins react with iron. We saw the ferric chloride test. So when you put it in an iron vessel, there are good chances that the tannins will try to chelate and react, forming complexes with the iron metal. This happens, and as a result, you will get a slightly darker shade compared to your previous example, which is pale catechu.

So this is done for enough time if you want to improve the color. You know, people often try to pass off black catechu as pale catechu. If you want to enhance the quality or the color of it, what they do is keep it in a glass-lined vessel where the interactions with the surface are minimal. So in that case, or even with stainless steel vessels, you will

you will get a much lighter shade of catechus almost matching to that of your pale catechu. So that is done. When the extract is transferred, cooled, and then allowed to set. This is generally set in wooden trays where it almost after keeping overnight solidifies. Once it solidifies, it's cut into pieces and sold in market as your cutch or catechu.

Now this is there with your panwalas and you will see it as nice cubes or blocks. Now what it contains is as compared to pale catechu which had about 7 to 30 percent of catechin. Here the catechin content is much less. So you will see 2 to 12 percent catechin again depending upon the source. So this is your catechin content.

And if you see this bond particularly, if you change it, you get what is called epicatechin. So you have catechin and epicatechin as monomers. If you remember, these are pseudotannins. They will polymerize to form phlobatannins, which are called catechutannic acid. So about polymerized particles or polymerized tannins, you will see about 25 to 33%.

Now, compare that to the pale catechu. Pale catechu had 25 to 55 percent of phloba tannins, especially catechu tannic acid. Now, apart from that, a little difference here is it doesn't contain your gambel tannin or gambel fluorescent. But here you have more of this acacatechin, especially the DL-acacatechin and iso acacatechin derivatives. In addition to that, this is more rich in gummy matter and you know that gives you a good blend or good taste.

Coming to the chemical test, if you see because it is lacking in gambier fluorescence, the black catechu will not give you a fluorescence test. because it is not prepared from twigs, it will not have chlorophyll. So these two tests are absent in black catechu, whereas the general test for condensed tannins, if you remember, it will obey your vanillin hydrochloric acid test

and it will also obey your matchstick test because it has catechin. The test unique to black catechu is if you treat a little bit of this black catechu with lime water, It will form a complex which is more brownish. And if you keep it standing for a period of 5 to 10 minutes, this brownish color will turn into a reddish color precipitate. So it will form a colloidal complex which will be more reddish and on standing.

The second one which is more unique here is if you see your condensed tannins. They give you a greenish coloration with ferric chloride. So with iron salts, even your black catechu gives you a green color. But this time the iron salt is ferric ammonium sulfate. Now this if you add sodium hydroxide.

So if you add sodium hydroxide to this, that green color gradually turns into a purplish hue. So these are two tests which will help you tell whether it's black catechu or not. Now, applications: black catechu is very medicinal, and that's the reason it's given as an astringent and as an antidiarrheal. It is known to cure diseases of the mouth and certain bacterial infections, and that's the reason it was actually added to paan.

So if you see the panwala applying black catechu, it's actually medicinal, and its astringent properties kill bacteria and help fight mouth odor. Now, one more interesting phenomenon if you observe here is that the catechu reacts with the lime. As well as the areca nut, which we refer to as supari in paan, it gives an interesting red color complex, which you will find associated with people chewing paan.

They often spit a reddish coloration. Now, this is actually a chemical test formed due to complexation. Apart from that, being a tannin, it is used for dyeing fabrics and gives a nice brown-black coloration. So here are a few references from the textbook which you can read too. And thank you, everyone, for your patient listening.

Thank you.