



COFFEE INDUSTRY CORPORATION LTD

COFFEE RESEARCH INSTITUTE

P.O.Box 105
Kainantu 443
Eastern Highlands Province
Papua New Guinea.

Telephone 7373511
7373518
7373552

Coffee Wastes, Soluble Dietary Fibre, Antioxidants and Health.

sdf8.wpd/kcc
Oct. 1999.

Ken C Calvert.
Prin.R.S. Processing.

A search on the Internet under the heading of “Dietary Fiber” (American spelling of course) brings a plethora of hits under a whole range of heads. Try ‘Soluble Dietary Fiber’, and the number of hits is reduced by about 20,000, but the spread of perceived properties and proven benefits actually increases. The intention of this paper is to point out that coffee mucilage, and cherry pulp, presently treated as waste products and highly polluting ones at that, are also a wonderful and natural mix of most of the prevalent terminology in the alternative health industry today. Those factors are all wrapped up into a series of potentially valuable products which could turn around the present economics of the plantation coffee industry. We are talking about lipid lachers, cholesterol absorbents; heart protectants, cross linked mineral carriers, antioxidants, bioflavonoids, oligo-proanthocyanins, gentle laxatives, anti aging and anti-cancer agents. However, these are the current buzz words of not only the alternative ‘Granola nuts’ but also the genuine medical fraternity, as demonstrated by many of the references gleaned from authentic medical journals.

We are of course also talking about alkaloids, tannins and other plant protection chemicals and antinutritional factors which have long prevented the free use of coffee pulp as an animal feedstuff at anything more than 10% of the basic diet. The change of paradigm however has come not only from new work on soluble dietary fibre and antioxidants, but also from the realization that the difference between a tonic and a poison is often only a matter of degree, and nobody has looked at the benefits of coffee pulp at less than 10% of the diet. Furthermore, bypassing the high levels of crude fibre in coffee pulp by methods of solution and extraction, just as we do with the traditional treatment of coffee beans, those positive nutritional factors can be enjoyed by the monogastric Homo sapiens, as well as those ruminant animals for whom the coffee cherry has originally evolved.

WHAT ABOUT COFFEE MUCILAGE!!!

Here at the PNG Coffee Research Institute in Aiyura, some very simple methods have been worked out to extract some interesting materials, not only from coffee mucilage but also from fresh coffee pulp and waste water. Given a few clues, and they are freely available in the literature, most chemists would soon be able to duplicate this work. In the past, much work has been done to extract pure food chemicals like pectin, caffeine, chlorogenic acids and enzymes from coffee wastes. In this pure form, chemicals such as pectin have to compete on world markets with lots of more concentrated and profitable sources like citrus albedo and apple pomace. What has been done here in the first instance is to look at these materials as natural unrefined specialty health supplements which are priced in a completely different fashion to basic foodstuffs.

From coffee industrial wastes we can obtain the following types of materials in varying states of purity;

A/. Unrefined pectins, (ie.crude) soluble dietary fibre or SDF, mainly from the mucilage.

These pectins can be either thermo reversible soluble gels or non reversible crosslinked ones which have a different mouth feel. The cross linked type is insoluble and so can be soaked to diffuse out the off flavours to give a very bland product. The thermo gel dissolves in water and so cannot be soaked as such. However it can be precipitated out of aqueous solution with alcohol, and purified to the level desired to contain the required amounts of antioxidants.

B\.. Natural coffee fruit sugars, mainly from the recycled pulping water. These contain very little sucrose. They are mostly monosaccharides, glucose, galactose, rhamnose and arabinose, with a different flavour, reminiscent of plums, and could be marketed as something new for the more sophisticated coffee connoisseur. The present problem is that many of these sugars are attached to both the flavonoids and the pectins present, and although they can be easily split off with dilute acid and heat, it is difficult to get a clean white product. That may or may not be a good thing. 'Coffee crystals coloured ala coffee.'

C\.. A collection of antioxidants and flavonoid compounds, mainly from the skins but also some from the de-esterified mucilage. These are mainly the anthocyanin fruit colour compounds, but they also contain all the other polyphenolics such as the chlorogenic acids and of course the caffeine. These materials can be put together into several combinations to make a range of food additives which should be of interest to the 'health food' industry. I say 'health food' or 'health supplement', because being natural products from a well known commodity, these materials could be test marketed almost immediately through this sector of the market economy and their popular appeal analyzed on the specialty market before anyone has to commit themselves to the heavy capital investment of large scale industry. A search on the Internet using the terms 'proanthocyanidins' or 'leucoanthocyanidins', brings up a plethora of pills and potions made from pine bark or grape seed extracts or a multitude of other things. All of these are priced in the range of pharmaceuticals and not as drinks, fruit extracts or materials for inclusion in basic foodstuffs. The great majority of authentic research on these materials has looked upon them as colours rather than flavours, and has been caught up in the intricacies of their chemistry at varying pH levels. Past references to their nutritive value are few and far between. However, with the spiraling interest in alternative medicinals, that situation is rapidly changing.

D/. One alternative refinement to C/. above, would be to extract these same colour chemicals, including the similar but colourless pro anthocyanins, and then freeze dry or crystallize out the concentrates as a resource base for other food manufacture or perhaps the more sophisticated synthesis of other chemicals. The production of 'enocianina' from red grape pomace is more than 100 years old. (Markakis 1982). The major problem with enocianina however is the sulphur dioxide content used in its extraction. This can give some unpleasant allergic reactions. A recent move has been with B-lycopene from tomatoes. (Tucker & Grierson 1987).

It is my personal conviction that the research on how to stabilise anthocyanin food colours by co-pigmentation,(Osawa 1982; Brouillard 1982), or by acylation or diacylation, (Ribereau-Gayon.1982;) is a real possibility for the production of stable food colours from coffee pulp. The thing is that most of these anthocyanin components of coffee pulp are already linked to Caffeic Acid, Chlorogenic and Dichloroquinic acids etc., which Timberlane & Bridle. (1982) indicate as the best acylating agents. Furthermore, the main colour compound is a prodelphinidin (Clifford et al.1991.), and is particularly noted for its ability to be diacylated. (Timberlake & Bridle 1982). Concentrating these chemicals by simple evaporation or just boiling does lead to a certain amount of condensation and formation of tannins. However, it does appear that rapid freeze drying can provide a more stable material, which will keep in the dehydrated form. This could be used to create both the colour and the flavour for the production of a health fruit drink powder such as 'Tang' does with orange juice..

E/. The food industry today is finding that the traditional commercial synthetic antioxidants such as BHA and BHT are no longer acceptable, and they are looking for a new range of 100% natural products to protect processed foodstuffs from rancidity and off flavours etc. (Amiot et al.1997). Anti oxidants come as 'in vivo' and 'in vitro' types. The 'invitro' ones are largely required to be fat soluble and are used for protecting cooking oils and fats etc. and need to be colourless and tasteless. The ones meant for dietary purposes are largely water soluble and can come in any flavour or colour. The fruitier the better. Coffee wastes are a good source of the latter but do also have a range of colourless flavanoid chemicals which could be developed for use as oil and fat protectants..

Whether or not these compounds can be classified as'Oligo Proanthocyanidins' or OPCs, and correspond to the much debated Vitamin P, or to 'Co-Vitamin C' is not known at this stage. (Schwitters 98) Nevertheless, they would be a good starting point for further research. And, being essentially glycosolated cyanins rather than cyanidins, all the current patent battles on OPCs can be circumvented.

The thing is that a tonne of waste coffee pulp costs a lot less than a tonne of tomatoes or of Grape pomace. And, there are not many tomato factories around that take in upwards from 20 tonnes a day very day for 5 months of the year and never actually close. This process would be the cheapest of all to implement and provide a valuable alternative income for the most hard pressed sector of our economy.

From a National and Industry point of view the extraction of these materials could also substantially reduce the waste water and pollution problems from our largest factories. These of course are the ones with real pollution problems as far as our environment is concerned, not to

mention the Govt Dept. of Conservation and Environment, and they would be the obvious sites to start on this kind of work. One opinion has estimated the wholesale value of these by products at around K2,000-00/tonne of green bean, (US600-00), at the usual profit margins. That sort of money could give a real fillip to the hardest pressed sector of the coffee economy. Furthermore, an additional monetary incentive to pick and process only ripe coffee cherry would do wonders for the overall quality of our top lines of coffee. Once having these base chemicals extracted, the residues can still be composted and used for organic fertilisers and soil improvers, with little loss of their preceived properties.

The overall thrust of this document is to suggest that most coffee producing countries have neither the infrastructure nor the expertise to either produce or market many of the above more sophisticated suggestions. However, we do have the ability to produce large quantities of semi refined raw materials from coffee wastes for export sale to pharmaceutical manufacturers and health food companies in the more sophisticated countries, for processing and marketing under their own brands. Most of those sort of companies however, would want to jump on the 'coffee' bandwagon to advertise their products, and so some sort of alliance between them and a legitimate coffee marketing interest may be required. The establishment of such industry will take a lot of time, and even more money. In this interim period, we do however have the ability to produce a number of simple materials, as elaborated below which could be used to produce an interim cash flow and to publicise these new coffee products.

SOME PRACTICAL MATERIALS:

The simplest material consists of the fermented mucilage fraction from the beans, blended back in with the condensed pulping waters from ripe cherry, which contain most of the natural sugars. Depending on how the mucilage is de-esterified, this mix when boiled down after the manner of maple syrup, dries into either a light brown sweet chewy substance called for the moment 'Coffee Leather', or to a darker and more fibrous material akin to liquorice. Despite its sweet taste, Coffee liquorice or perhaps, 'Coffee Toffee' does have a distinctive flavour and it is of course more of a laxative than a sweet, with gentle medicinal and prophylactic properties akin to the said liquorice. This could be marketed immediately as a health supplement and a pleasant way of increasing ones intake of both soluble dietary fibre and antioxidants. At present it is not known how much caffeine these materials contain, but it is much less than we might surmise. Initial production rates for Coffee Toffee are around 300gms. per 12 litres of heavily recycled pulping water.

It should be pointed out that these materials, like liquorice, do not look nice in the raw. In order to avoid such a raw grey fibrous appearance, liquorice is always extruded in thin straps or rods through polished dies with lubricant to give a polished black appearance on the surface and a minimal cross section to expose the cruddy interior. This also aligns the fibres, but of course appearance is everything. This would also be the approach to market these substances.

The next more complex type of material is straight mucilage, stripped with acid enzyme, the sugars disconnected, to make cyanidins rather than cyanins, and with the pectin fraction partially cross linked with calcium or iron salts, deionized and then dried at acid pH. To get a gel rather than a solid mass of syneretic co-aggregate some care is required. To avoid syneresis, it is necessary to add the calcium under boiling conditions, and let the gel form on cooling. (Pilnik & Voragen 1970) This gives a pale coloured gelatinous substance which I have yet to have freeze

dried (problems with our freeze dryer), but which I believe will come out as a crystalline substance akin to 'Metamucil'. Hopefully this can be used not only as a sweetener with normal coffee but also as a source of soluble dietary fibre. Depending on the degree of cross linking the gel is either thermo reversible or insoluble.

This means that a couple of spoons of the thermo reversible stuff stirred into hot coffee will give a cup with the same traditional flavour but also with a lot of body and a distinctly different mouth feel. If the coffee is allowed to cool it will gel in the cup, the acme of good soluble dietary fibre. It is my feeling that this product would appeal to the Yuppy sector, (Young urban professionals), many of whom are reputed to down a cup of black coffee on the way out the door in the morning and hope to stay slim by eating nothing until 10zs' or even until lunch. A somewhat gimmicky aspect is that if one chews up a couple of calcium based antacid tablets to maybe counter the gastric irritation problems of the malic acid, then the pectin will cross link even further in the stomach and become very filling indeed. Such calcium cross linked gels are the basis of the whole 'diet' tinned fruit industry. Ferrous ions will do it better, but there are colour and flavour changes involved.

The third simple possibility is to acid extract the red colour compounds of the skins and then concentrate this along with sugars, preferably from the sugary recycled pulping waters, to get a deep red colored cordial extract, 'Cherry Coffee Syrup', with its own distinct flavour. This could be advertised, unlike most cordials, as all natural, no artificial colours no artificial flavours and what is there is like a tonic and very good for you! The flavour is sharp, because of the acid needed to keep the colour bright red, (pH3.0, like Coca Cola). The taste is akin to blackcurrant juice and at times has the same after taste. I am still working on this to understand and try and eliminate that problem by means of ion exchange. Because the colour is a bright red, sometimes bluish red, it might be even more appealing than dark grape juice but with all the same antioxidant properties of red wine. No alcohol, but caffeine instead! It could also be freeze dried to a reddish powder, like 'Tang', and which should have similar properties. Such an approach would minimize the condensation and precipitation of the colour as a brown sediment, such as does happen with red wine and most anthocyanins. However, beyond its basis as a natural fruit extract, the actual antioxidant qualities, caffeine content etc., would have to be proven by chemical analysis.

WHERE DO THE HEALTH ASPECTS COME FROM?

In brief, the medicinal properties of these materials can be described in the following terms

1/. Soluble Dietary Fibre and Heart Disease.

Atherosclerosis, is the loading of our arteries with deposits of cholesterols, (ie. low density lipoproteins or LDLs). Of prime importance are our coronary arteries and the dangers of a heart attack. A few years ago everybody was rushing to eat oat fibre, because it was shown to absorb LDLs out of our blood plasma. Experts are now telling us that pectins are better than oat fibre (Johnson & Southgate 1994; Fourie 1996), because as well as reducing the LDLs, pectins also boost the levels of the high density HDLs, which are the really beneficial ones.

Pectins are well known for locking up bile acids, (where those cholesterol come from), and taking them on through the small intestine to the colon or large intestine, where some of them become food for bacteria which in turn protect against colon cancer. (Hill 1984). The rest are excreted as tannin complexed non digestibles, and thus lower the overall cholesterol levels. The new part of the story however, is the role of short chain fatty acids, (SCFAs), in protecting against colitis and diverticular disease as indicated in the appended editorial from the American Journal of Gastroenterology.(Floch 1990), SCFAs are produced by bacterial action on soluble dietary fibre in the colon.

2/. Cation exchange properties.

Pectins, in the form of galacturonic oligosaccharides, are a bit like ion exchange resins. (Furda 1979, Blaney 1996.). They are able to complex with free calcium, iron and other divalent metal ions in the diet and carry them out of the body, seriously reducing the levels of these important nutritional constituents. This could lead to osteoporosis and anemia. However, a possible turnaround in this situation would be to prestabilise the pectinacious material by cross linking it with either or both of these metals before it is ingested. (Which is what we have done.) Pectins might then become powerful dietary agents to control these diseases as they drop some of their metal ions in the acid conditions of the stomach, in order to pick up the lipo-proteins later on in the classical cation exchange reaction. (Eastwood 1991) That would be a win win situation. In the case of iron it is also necessary to protect it through the alkaline conditions of the duodenum and avoid it being precipitated as insoluble hydroxides and then pass right through the body without being absorbed at all. Once again pectins are of great value in achieving this.

3/. Antioxidants.

Coffee mucilage, but more particularly the pulp, is not all pectins or protopectins. It also contains a number of linked sugars and polyphenolic chemicals, anthocyanins, proanthocyanins, and cyanidins, bioflavonoids and tannins, not to mention caffeine and chlorogenic acids. (Stadler 1995, Vinson et al 1995, Millar 1996). In the past, when all that chemists were thinking about was the production of pure food component chemicals, these were considered to be the main contaminants, to be removed. Now, these compounds are very much in vogue as scavengers of 'free radicals'. In particular, hydroxyl, peroxy, and singlet oxygen radicals. The papers appended, (Anon 1997, Davasagayam et al. 1996), point out that these polyphenolic antioxidants are much superior to Vitamin C and Vitamin E which are presently in vogue for their ability to bind free oxygen. These free radicals are reputed to be a major factor in aging as they cross link muscle fibres to cause stiffness and also damage our DNA to cause a number of aging diseases, including some cancers. These problems can now be partially controlled by coffee chemicals (Nagasawa 1995). The fact that these chemicals are all part of a completely natural product with no synthetic additives and the only genetic modifications happened millions of years ago, constitutes another win win situation.

It should of course be pointed out that most of these benefits are also conferred by eating lots of fresh fruit. "An apple a day---", particularly ones with red skins, will supply many of these chemicals. So there is nothing new, its just a case of getting these components together with the right kind of sales strategies and in a pleasant way to eat. How many people peel their apples,

before eating, because they don't like the part where most of the benefits reside?

4/. Even Caffeine is losing its bad image.

Caffeine and more particularly those chlorogenic acids are particularly good antioxidants.(Stadler 1995, Vinson et al 1995, Millar 1996). Q.V also the news clip, 'a cup of coffee is equal to three oranges'.(Anon.1997). Just note that all those references are less than five years old.

5/. Fat Replacer.

An already well established technology is the use of pectin emulsions to replace fat emulsions in cooking and the manufacture of salad dressings and mayonnaise. A short paper on a proprietary formulation is appended as an indication of the uses of this material.

6/. The Chemical composition of coffee wastes.

The mucilage fraction.

According to the best sources, (Brehm & Bressani 1979;), quote the wet composition of coffee mucilage as,

- 84.2% water.
- 8.9% protein. (Including polyphenolics.)
- 4.1% sugar.
- 0.91% pectic acid.
- 0.7% ash.

They also state, from another source, that dehydrated coffee mucilage represents about 5% of the dry weight of the coffee cherries. It consists of;

- 35.8% pectin substances.
- 45.8% sugars.
- 17.0% cellulose and ash.

The sugar fraction is 60% reducing sugars and 40% non reducing sugars. There is little if any caffeine in the mucilage. From the obvious divergence of these figures and their age, >20 years, it is obvious that, despite the immense amount of research that has been on green coffee beans and the flavours developed after roasting, there is a wide open gap around fresh coffee materials which cannot be easily transported to an overseas laboratory. The Coffee Industry needs to promote the analysis of fresh coffee wastes in the light of current trends and ideas.

The polyphenolic fraction.

The most recent work here has been done by Clifford, Ramirez-Martinez and de Menezes According to Ramirez (1987.), there was identification of compounds by paper Chromatography within the following groups. Chlorogenic acids, Catechins, leucoanthocyanins, anthocyanins, and flavonol. glycosides. Clifford and Ramirez-Martinez (1991), in a more detailed study found from 0.40-1.61% of caffeoylquinic acids and 0.54-1.67% caffeine in the dried pulp. Most work has been done on biological fermentation studies, to try and reduce the toxicity and allow the use

of pulp as an animal feedstuff. (Rolz.88, Menezes et al 93&95, Ramirez-Martinez 98.) Despite some other such detailed sections of analysis, (Clifford et al 1992, 93, 94.), there is no wide ranging study with a broad analysis of coffee pulp wastes.

7/. Assessing the negatives.

When looking at antioxidants in general, some of the polyphenolic antioxidant chemicals, usually quoted as protein or nitrogen, do have negative biological properties. After all, the main properties of tannins, as presently indicated, are as plant protection agents, locking up proteins from attack by insects, animals and microorganisms. (Harbourne 1997,1&2). However, in the appended papers on this subject, (Madhavi & Salunke 1996, Chung & Wei 1997, Barlow 1990, Bermond 1990.), the main pathogenic components mentioned are either man made synthetic chemicals used in large scale food manufacturing, like BHA and BHT, or the older traditional leather tanning agents, altogether the most vicious end of a wide span of compounds coming mainly from bark, leaves and wood. They are certainly not from fruit. (Harbourne 1997(2)). There is one reference to the finding that certain natural colours have been found to be mutagenic in vitro, notably the flavone derivatives kaempferol and quercetin which are widely distributed in vegetables and other plants (Drake 1980). Both of these compounds are found in goodly amounts in green coffee beans and will most likely be found in the pulp as well. The question will be as to whether they are largely eliminated in the roasting process or come through essentially unharmed into our favourite beverage.

Despite the work by Frischknecht (1986), showing that caffeine is not moved around the plant, The present hypothesis on the toxicity question, is that as the plant recycles the stronger protection agents out of its tender leaves when they mature and become less palatable, these protection agents are then subtly altered and recycled into its fruits as something more prophylactic and less toxic. The bronzing of new growth in some varieties of coffee is an additional visual warning of toxic tannin content. (Harbourne 1997(1))

The fact that certain parts of that cherry, like the mucilage, are very very difficult to breakdown, by anaerobic bacteria, which present theory indicates largely originated from the animal gut, just makes it all part of the pattern. The mucilage fraction of the coffee cherry cannot be made into biogas because it was designed to resist these same anaerobic bacteria.

Someone is also sure to ask the question as to how a reputedly dirty material that is only fit for running down the nearest drain can be used as a food material? The answer to that comes as an already established technology from Brazil. Because they make coffee from buni, of which much is swept up off the ground, there are coffee washing/sorting machines available which pre-process the cherries so that they come out as washed, water sorted and graded. That is, much like the apples, oranges and other fruits, not to mention root vegetables like carrots, that many people in developed economies love to buy and juice or eat raw every day. Environmental Pulpers will surely become the future for coffee processing, because they do not leach the solubles out of the pulp.

8/. Can we afford to be involved?

One may well ask the question, why should a poor third world country start mixing it in with the big boys running research institutions in Japan, Europe and the USA? After all that is where most of the appended research papers have come from. The whole health food supplements industry is concentrated in the consumer countries. One should also note however, that all that past work on coffee wastes has been done on dried and preserved samples. Europe, Japan and the USA do not grow coffee, and probably the reason that they

have not done any work on raw unrefined mucilage, far superior to anything else for this kind of work in prophylactics is the fact that they have never seen it. Dried cherry solids, as distinct from mucilage solids, do not react in the ways indicated above. This fact also minimises the effort of most of those Research Institutes in Latin America, where buni coffee production is dominant. So, even in 1999, there is still room for third world coffee producing countries to be involved in ground breaking developments. However, we should concentrate on producing the raw materials in bulk, in a stabilised form, for onward sale to all those big Multinationals who can afford to refine and market the whole range of retail products each in their own corners of the world.

9/. Why do it in P.N.G?

If we want to boost our international image, not only as one of the top coffee countries in the world but also as being on the cutting edge of R.&D., then there is only a small window of opportunity to do a bit of basic work on these products and set the stage with a patent or two, and then stand aside for the rush. At the present moment Papua New Guinea stands poised at the top of the 'Other Milds' bracket. The ground breaking development of a new sphere of by products for the coffee industry, such as outlined above would surely tip the balance. That balance would project not only Papua New Guinea, but also every other third world coffee producer, into a new era of value added production of commodities which have to be dealt with and processed on the spot rather than dried and exported as basic raw materials to avoid punitive tariff protection. At the moment the coffee producing countries are squeezed into exporting green bean as a basic commodity and all the value adding is done overseas. For the future of the third world that trend needs to be reversed.

oooOOOoo

REFERENCES:

- Amiot M.J., Fleuriet A., Cheynier V., Nicolas J., 1997.
Phenolic Compounds and Oxidative Mechanisms in Fruit and vegetables.
In, Phytochemistry of Fruit and Vegetables. Edit. Tomas-Barberan & Robins.
Oxford Science Pub. 1997.
- Anon. 1997.
Cup of Coffee Equivalent to Three Oranges.
Chemistry and Industry. No.8. Apl. 1997.
- Arjmandi B.H., Ahn J., Nathani S., Reeves R.D., 1992.
Dietary Soluble Fiber and Cholesterol, Affect Serum Cholesterol Concentration, Hepatic Portal Venous Short-Chain Fatty Concentrations and Fecal Sterol Excretion in Rats.
American Institute of Nutrition. 0022-3166/92.

- Barlow S.M., 1990.
Toxicological Aspects of Antioxidants used as Food Additives.
In, Food Antioxidants. Edit. B.J.F. Hudson.
Elsevier. App. Science series. 1990.
- Bermond P., 1990.
Biological Effects of Food Additives..
In, Food Antioxidants. Edit. B.J.F. Hudson.
Elsevier. App. Science series. 1990.
- Blaney S., Zee J.A., Mongeau R., Marin J., 1996.
Combined effects of various types of Dietary Fiber and Protein on in Vitro Calcium Availability.
J.Agric. Food Chem. **44**.3587-3590. 1996.
- Bradbury A.G.W., Halliday D.J., 1990.
Chemical Structures of Green Coffee Bean Polysaccharides.
J.Agric. Food Chem. **38**. 389-392. 1990.
- Braham J.E., Bressani R., 1979.
Coffee Pulp: Composition, Technology, and Utilization.
IDRC. 1979.
- Braudo E.E., Soshinsky A.A., Yuryev V.P., Tolstoguzov V.B., 1992.
The Interaction of Polyuronides with Calcium Ions. 1: Binding Isotherms of Calcium Ions with Pectic Substances.
Carbohydrate polymers. **18**. 165-169. 1992.
- Brouillard R., Figueiredo P., Elhabiri M., Dangles O., 1975.
Molecular Interactions of Phenolic Compounds in Relation to the Colour of Fruit and Vegetables.
In, Phytochemistry of Fruit and Vegetables. Edit. Tomas-Barberan & Robins.
Oxford Science Pub. 1997.
- Brouillard R., 1982.
Chemical Structure of Anthocyanins.
In Anthocyanins as Food Colours. Edit. Markakis P.
Academic Press. 1982.
- Chang M.L.W., 1983.
Dietary Pectin: Effect on Metabolic Processes in Rats.
In; Unconventional Sources of Dietary Fiber.
A.C.S. Symposium 212. 1983.
- Cheynier V., Moutounet M., 1992.

- Oxidative Reactions of Caffeic Acid in Model Systems containing Polyphenol Oxidase.
J.Agric. Food Chem. **40**. 2038-2044. 1992.
- Chun W., Bamba T., Hosada S., 1989.
Effect of Pectin, a Soluble Dietary Fiber, on Functional and Morphological Parameters of
the Small Intestine in Rats.
Digestion. **42**.22-29. 1989.
- Chung K.T., Wei C.I., 1997.
Food Tannins and Human Health: A Double-Edged Sword.
Food Technology. Vol. 51. No.9. Sept 1997.
- Clifford M.N., 1986.
Phenol-Protein Interactions and their Possible Significance for Astringency.
In, Interactions of Food Components. Edit. Birch G.G., & Lindley M.G.,
Elsevier. 1986.
- Clifford M.N., Ramirez-Martinez J.R., 1991.
Tannins in Wet-processed Coffee Beans and Coffee Pulp.
Food Chem. **40** 191-200.
- Clifford M.N. Gonzalez de C.N., Ramirez-Martinez J.R., Adams M.R., de Menezes H.C., 1992.
Tannins in the Sun Dried Pulp from the Wet-Processing of Arabica Coffee Beans.
ASIC. Paris. 1992.
- Clifford M.N., Gonzalez N.de C., Ramirez-Martinez J.R., Aldana J.O., 1993.
Progress in the Analysis of Proanthocyanidins in Freshly Prepared Coffee Pulp.
ASIC. 15th Coll. 1993.
- Clifford M.N. 1997.
Astringency.
In, Phytochemistry of Fruit and Vegetables. Edit. Tomas-Barberan & Robins.
Oxford Science Pub. 1997.
- Colmenares N.G. de, Ramirez-Martinez J.R., Aldana J.O., Clifford M.N., 1994.
Analysis of Proanthocyanidins in Coffee Pulp.
J.Sci.Food & AGRIC. **65**, 157-162. 1994
- D,Mello J.P.F., 1997.
Toxic Compounds From Fruit and Vegetables.
In, Phytochemistry of Fruit and Vegetables. Edit. Tomas-Barberan & Robins.
Oxford Science Pub. 1997.
- Devasagayam T.P.A., Kamat J.P., Mohan H., Kesavan P.C., 1996.
Caffeine as an Antioxidant: Inhibition of Lipid Peroxidation Induced by Reactive Oxygen

- Species.
 Biochemica et Biophysica Acta. 1282 (1996) 63-70.
- Drake J. J-P, 1980.
 Toxicological Aspects.
 In, Developments in Food Colours-1.
 Edit. J.Walford. Applied Science Pubs. 1980.
- Eastwood M.A., 1991.
 Physical Properties of Fibre Towards Bile Acids, Water and Minerals.
 In; Dietary Fibre. Edit. Birch G.G., Parker K.J.,
 Academic Press. 1991.
- Fleming S.E., Calloway D.H., 1982.
 Determination of Intestinal Gas Excretion.
 In; Dietary Fibre. Edit. Birch G.G., Parker K.J.,
 Academic Press. 1982.
- Floch M.D., 1990.
 Soluble Dietary Fiber and Short Chain fatty Acids: An Advance in Understanding the
 Human Bacterial Flora.
 American Journal of Gastroenterology.
 Vol. **85**. No.10. 1990.
- Fourie, P.C., 1996.
 Fruit and Human Nutrition.
 In, Fruit Processing. Edit. Arthey D., Ashurst P.R.,
 Blackie. A.&P. 1996.
- Frischknecht P.M., Ulmer-Dufek J., Baumann T.W., 1986.
 Purine Alkaloid Formation in Buds and Developing Leaflets of Coffea Arabica:
 Expression of an Optimal defence Strategy.
 Phytochemistry. Vol.25. No3. 613-616. 1986.
- Furda I., 1979.
 Interaction of Pectinaeous Dietary Fiber with some Metals and Lipids.
 In, Dietary Fibers, Chemistry and Nutrition.
 Academic Press. 1979.
- Garcia R., Arriola D., de Arriola M.C., de Porres E., Rolz C., 1991.
 Characterization of Coffee Pectin.
 Lebensm.-Wiss.- Technol., **24**, 125-129. 1991.
- Harbourne J.B., 1997(1)
 Role of Phenolic Secondary Metabolites in Plants and their Degredation in Nature.
 In, Driven by Nature: Plant Litter Quality and Decomposition. Edit. Cadish & Giller..
 CAB International 1997.

- Harbourne J.B., 1997.(2)
Phytochemistry of Fruits and Vegetables. An Ecological Review.
In, Phytochemistry of Fruit and Vegetables. Edit. Tomas-Barberan & Robins.
Oxford Science Pub. 1997.
- Hill M.J., 1984.
Bile Acids and Colorectal Cancer in Humans.
In; Dietary Fiber. Basic and Clinical Aspects.
Edit. Vahouny & Kritchevsky. Plenum Pub. N.Y. 1984.
- Ito M., Deguchi Y., Matsumoto K., Kimura M., etal. 1993.
Influence of Galacto-oligosaccharides on the Human Fecal Microflora.
J.Nutr.Sci.Vitaminol., **39**. 635-640, 1993.
- Jensen C.D., Haskell W., Whittam J.H., 1997.
Long-Term Effects of Water-Soluble Dietary Fiber in the Management of
Hypercholesterolemia in Healthy Men and Women.
Amer. J. Cardiol. **79**. 34-37. 1997.
- Johnson I.T., Southgate D.A.T., 1994.
In Dietary Fibre and Related Substances.
Chapman & Hall. 1994.
- King B.M., Solms J., 1982.
Interactions of Volatile Flavor Compounds with Propyl Gallate and other Phenols, As
Compared to Caffeine.
J.Agric. Food Chem. 1982, 30, 838-840.
- Klaur H., Pongracz G., 1981.
Ascorbic Acid and Derivatives as Antioxidants in Oils and Fats.
In, 'Vitamin C.' (Ascorbic acid.) Edit. Counsell J.N. & Hornig D.H. 1981
- Kritchevsky D., 1984.
Fiber and Cancer.
In; Dietary Fiber. Basic and Clinical Aspects.
Edit. Vahouny & Kritchevsky. Plenum Pub. N.Y. 1984.
- Liener I.E., 1980.
In; Toxic Constituents of Plant Foodstuffs. Chap.1.
Edit. Liener I.E. Academic Press. U.K. 1980.
- McBurney M.I., Horvath P.J., Jeraci J.L., Van Soest P.J., 1985.
Effect of in Vitro Fermentation using Human Fecal Inoculum on the Water-Holding
Capacity of Dietary Fibre.
Brit. Journal of Nutrition. **53**. 17-24. 1985.
- Madhavi D.L., Salunkhe D.K., 1996.

- Toxicological Aspects of Food Antioxidants.
In, Natural Antioxidants and Food Quality in Atherosclerosis and Cancer Prevention.
Edit. Kumpulainen J.T., & Salonen J.T. Cambridge Press 1996.
- Markakis P., 1982.
Anthocyanins as Food Additives.
In Anthocyanins as Food Colours. Edit. Markakis P.
Academic Press. 1982.
- Menezes H.C., Samaan F.S., Clifford M.N. Adams M.R., 1993.
The Fermentation of Fresh Coffee Pulp for use in Animal Feed.
ASIC. 15th Coll. 1993.
- Menezes H.C., Romeiro L.R., 1995.
The Incorporation of Coffee Pulp in Animal Feed.
Reduction of Growth Impairment by Fermentation.
ASIC. 16th Coll. 1995.
- Millar N.J., 1996.
The Relative Antioxidant Activities of Plant derived Polyphenolic Flavonoids.
In, Natural Anti oxidants and Food Quality in Atherosclerosis and Cancer Prevention.
Edit. Kumpulainen & Salonen. Cambridge Press 1996.
- Nagasawa H., Yasuda M., Inatomi H., 1995.
Protection by Coffee Cherry of Development and Growth of Spontaneous Mammary
Tumours in Mice.
ASIC. 16th Coll. Kyoto, 1995.
- Osawa Y., 1982.
Copolymerization of Anthocyanins.
In Anthocyanins as Food Colours. Edit. Markakis P.
Academic Press. 1982.
- Pilnik W., Voragen A.G.J., 1970.
Pectic Substances and other Uronides.
In, The Biochemistry of Fruits and their Products
Academic Press. 1970.
- Porter L.J., Hrstich L.N., Chan B.G., 1986.
The conversion of Proanthocyanidins and Prodelphinidins to Cyanidin and Delphinidin.
Phytochemistry, Vol.25. No1. 223-230. 1986.
- Pratt D.E., Hudson J.F., 1990.
Natural Antioxidants Not Exploited Commercially.
In, Food Antioxidants. Edit. Hudson B.J.F.
Elsevier. App.Food Science Series. 1990.

- Pszezola D.E., 1991.
Pectin's Functionality finds use in the Fat-Replacer Market.
Food Technology. Dec. 1991.
- Rajalakshmi K., Narasimhan H., 1989.
Food Antioxidants; Sources and Methods of Evaluation.
In; Food Antioxidants; Technological, Toxicological, and Health Perspectives.
Edit. Madhavi, Deshpande, Salunke. 1989.
- Ramirez-Martinez J.R., 1987.
Compuestos Fenolicos en la Pulpa de Cafe. Cromatografía de Papel de Pulpa Fresca de 12
Cultivares de Coffea arabica L.
Turrialba Vol. 37, No4. 317-323.
- Ramirez-Martinez J.R., 1988.
Phenolic Compounds in Coffee Pulp: Quantitative Determination by HPLC.
J.Sci. Food & Agric. **43**. 135-144. 1988.
- Ramirez-Martinez J.R., 1998.
Coffee Pulp is a By-Product, Not a Waste.
Tea & Coffee Trade Journal. April 1998.
- Rolz C., Menchu J.F., Espinosa R, Garcia-Prendes A., 1971.
Coffee Fermentation Studies.
ASIC. %th Coll. Lisbonne 1971.
- Rolz C., de Leon R., de Arriola M.C., 1988.
Biological Pretreatment of Coffee Pulp.
Biological Wastes. **26**. pgs 97-114 1988.
- Sakaguchi E., Sakoda C., Toramaru Y., 1998.
Caecal Fermentation and Energy Accumulation in the Rat Fed on Indigestible Oligo-
saccharides.
British Journal of Nutrition. **80**. 469-476.
- Schwitters H., 1998.
Why Proanthocyanins are the real Vitamin P and why their antioxidant capacity was
patented in the U.S.A.
www.inc-opc.com/
- Selvendran R.R., 1982.
The Chemistry of Plant Cell Walls.
In; Dietary Fibre. Edit. Birch G.G., Parker K.J.,
Academic Press. 1982.

- Stadler R.H., Fay L.B., 1995.
 Antioxidative Reactions of Caffeine: Formation of 8-Oxocaffeine (1,3,7-Trimethyluric Acid) in Coffee Subjected to Oxidative Stress.
 J.Agric. Food Chem. **43**. 1332-1338. 1995.
- Staub H.W., Mardones B., Shah N., 1982.
 Modern Dietary Fibre: Product Development and Nutrient Bioavailability.
 In; Dietary Fibre. Edit. Birch G.G., Parker K.J.,
 Academic Press. 1982.
- Theander O., 1982.
 Advances in the Chemical Characterization and Analytical Determination of Dietary Fibre Components.
 In; Dietary Fibre. Edit. Birch G.G., Parker K.J., 0
 Academic Press. 1982.
- Timberlane C.F., Bridle P., 1982.
 Distributuion of Anthocyanins in Food Plants.
 In Anthocyanins as Food Colours. Edit. Markakis P.
 Academic Press. 1982.
- Tucker G.A., Grierson D., 1987.
 Fruit Ripening.
 In, The Biochemistry of Plants. Vol 12. Pgs 265-318.
 Academic Press. 1987.
- Vinson J.A., Dabbagh Y.A., Serry M.M., Jang J., 1995.
 Plant Flavonoids, Especially Tea Flavonoids, are Powerful Antioxidants using an In Vitro Oxidation Model for Heart Disease.
 J.Agric. Food Chem. 1995. 43 2800-2802.

---oooOOOooo---