Fruit waste streams in South Africa and their potential role in developing a bio-economy

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Current and previous studies on bio-based (fruit) wastes and wastewaters, with a particular emphasis on research in South Africa, were reviewed. Previous studies have focused predominantly on the beneficiation and application of fruit waste as a feedstock for renewable energy. A definite gap in knowledge and application of fruit waste streams with regard to enzyme production as a value-added product is identified. The characteristics and composition of each type of fruit waste are highlighted and their potential as feedstocks in the production of value-added products is identified. The conversion of agri-industrial wastewaters to bioenergy and value-added products is discussed, with special mention of the newly published South African *Bio-Economy Strategy*, and the potential production of biofuels and enzymes from waste streams using recombinant *Aspergillus* strains. Finally, to maximise utilisation of waste streams in South Africa and abroad, a conceptual model for an integrated system using different technologies is proposed.

Introduction

Biorefineries and the bio-economy

For the past millennium, the world has run on crude oil and coal as the main energy source. In the past decade, the price of crude oil has doubled, and with climate change imminent, the world has to re-evaluate its economic growth and energy policies. South Africa, as part of Africa, has the added burdens of rising unemployment and poverty and the need to decouple its economy from fossil fuels. At the same time, the gap between the rich and poor is growing and food security remains high on the agenda. South African energy needs have been highly dependent upon abundant coal supplies; about 77% of South Africa's energy is directly derived from coal, with the balance stemming from nuclear power and hydroelectric resources. The demand to replace conventional industrial processes with those that generate fewer or no pollutants is increasing as a result of the need to minimise anthropogenic environmental impacts.[1] Our ability to meet market demands while maintaining environmental integrity is critically important for our future on earth.

Major considerations, with respect to the availability of renewable resources, as well as the appropriate technologies for converting these resources into the required commodities, are pivotal in the process of a societal transition to a bio-based economy.[2] Theoretically defined, a bio-based economy is an economy in which all inputs are derived from renewable resources.[3] The term 'bio-economy' was coined by the Biomass Research and Development Board in 2001, which used it to describe the revolutionary transition to a sustainable future by implementing a technology-driven model for economic development.[4]

In January 2014, the South African Department of Science and Technology revealed the national *Bio-Economy Strategy*.[5] In this document, the term bio-economy 'encompasses biotechnological activities and processes that translate into economic outputs, particularly those with industrial application'. The vision for South Africa sees the bio-economy contributing significantly to the country's gross domestic product by 2030 through the creation and 'growth of novel industries that generate and develop bio-based services, products and innovations'[5]. The potential of a thriving bio-economy will affect the country on a macro-economic scale, making South Africa internationally competitive (especially in the industrial and agricultural sectors) by creating more sustainable jobs, linking the countries first and second economies, enhancing food security and creating a greener economy.[5] The strategy presents a framework for the development of a thriving bio-economy, in which collaboration between role players (including, among others, the biotechnology sector as a whole, environmental agencies and social scientists) is the key to success. The three key economic sectors identified for inclusion in the strategy are agriculture, health and industry.[5] In order to sustain a future bio-based economy, methods for the conversion of renewable feedstocks into the respective value-added products will need to be efficient. Furthermore, there is a need to explore all possibilities in the use of sustainable resources to ensure the extraction of maximum value with minimum negative impact.

Accompanying the worldwide paradigm shift to environmental responsibility and sustainable development, there has been an increasing amount of research focused on developing technologies to produce or process biomass, for example, biofuel production, animal feedstock applications and extraction of value-added products. Generally, it is agreed that the development of biorefineries is crucial for the development of a bio-economy. The most inclusive of many definitions of a biorefinery was coined by the International Energy Agency Bioenergy Task 42 as 'the sustainable processing of biomass into a spectrum of marketable products and energy'[6]. The biorefinery concept neatly adheres to the ideals of a bio-economy, in which bio-based, renewable inputs are converted to valuable products using a wide range of technologies. It is imperative that negative environmental and social impacts are limited during these processes.

South African fruit industry

The long-term sustainability of biorefinery processes and products is reliant on a dependable supply of starting materials or substrates. The identification and quantification of potential input material is therefore a critical starting point in biorefinery design. This review is focused on the wastes generated from the fruit-processing industry in

South Africa. In order to be considered a useful, feasible feedstock, fruit wastes must:

- be produced in sufficient quantity (seasonality of the feedstock is an important consideration – see section on a conceptual model to maximise utilisation of fruit-waste streams for more information regarding this aspect) and
- have sufficient potential for value-addition, which outcompetes that of the current disposal method.

It must also be borne in mind that the carbohydrate content of most of the fruit-waste streams may be low, which also renders their commercial use a challenge (see section on a conceptual model to maximise utilisation of fruit-waste streams for more information on how this issue may be addressed).

The South African fruit industry produces a large variety of fruit, with citrus fruit, grapes, apples, pears, peaches and pineapples produced in the greatest quantities. A comparison of the amounts of fruits produced and processed in 2011/2012 is given in Table 1 (2011/2012 data).

Table 1: Production and processing data for various fruit crops in South Africa[7]

Fruit crop	Total production in tonnes (2011/2012)	Volume processed in tonnes (2011/2012)
Citrus (oranges, lemons, limes, grapefruit and naartjies)	2 102 618	441 899
Grapes	1 839 030	1649 (processed) 151 628 (dried) 1 413 533 (pressed)
Apples	790 636	244 469 (processed) 1110 (dried)
Bananas	371 385	Not indicated
Pears	346 642	120 811 (processed) 9872 (dried)
Peaches	190 531	125 706 (processed) 8994 (dried)
Pineapples	108 697	81 753
Watermelons and melons	93 277	Not indicated
Avocados	87 895	Not indicated
Apricots	66 762	48 792 (processed) 8725 (dried)
Mangoes[8]	65 439	∼50 000
Plums	60 925	1712
Guavas	23 699	20 896
Papayas	12 565	Not indicated
Litchis	7782	Not indicated
Strawberries	5543	2724
Other berries	5073	3914
Prunes	3426	Not indicated
Figs	1925	448
Pomegranates[9]	1324	883
Cherries[10]	775	83
Granadillas	484	Not indicated
Quinces	208	Not indicated

Note: processed = canned and/or juiced; dried = prepared as dried fruit; pressed = pressed for winemaking

South Africa comprises different temperate zones and fruit production is therefore scattered throughout the country. Production of the major crops – grapes, apples and citrus – is mainly centred in the Western Cape and Eastern Cape Provinces. In addition to deciduous fruit, sub-tropical fruit and other common fruit crops, South Africa also has a thriving olive industry, which is mainly based in the Western Cape Province. Presently, South Africans consume about 3.5 million litres of olive oil annually, of which local production only contributes 20%. The olive industry is one of the fastest growing agricultural sectors in South Africa with a growth rate of approximately 20% per annum. Olive production was estimated at 1500 tonnes for 2012/2013.[11]

Fruit processing (canning, juicing, winemaking and drying) generates large quantities of waste, both solid and liquid. For example, approximately 25–35% of processed apples (dry mass), 50% of citrus and 20% of grapes end up as waste.[12] The solid waste, often called pomace, is the portion of the fruit that is not utilised, such as skins, pips and fibres. The pomace has a high lignocellulose content and is very recalcitrant to degradation. In addition, large volumes of liquid wastes are generated from washing during processing. According to the *South African National Water Act of 1998*, wastewater must meet specified standards before it can be discharged into rivers or used for irrigation. Based on composition, there are limits to the volume of wastewater permitted for irrigation usage. For example, wastewater with a chemical oxygen demand (COD) of less than 400 mg/L can be used for irrigation at volumes of up to 500 m^3, while irrigation volumes may not exceed 50 m^3 on any given day if the COD is between 400 mg/L and 5000 mg/L. The average COD of wastewater in the juicing and canning industries is often as high as 10 000 mg/L and therefore requires extensive treatment before discharge into the environment.[12]

In a recent study by Burton et al.[13], it was recommended that maximum beneficiation of waste streams can be achieved through supplementation of the wastewater with solid waste, especially if the waste is targeted for microbial biomass or bioenergy production. South Africa produces sufficient fruit-processing wastes (solid and liquid) for the development of a biorefinery to be a viable option.

Composition and potential value of waste from selected fruits

Waste streams should be characterised to determine the potential for extraction of valuable products, microbial growth and/or enzyme production. The levels of nutrients need to be quantified to ascertain whether supplementation is necessary. Waste generated during the processing of an emerging crop (olives) and the major fruit crops produced in South Africa (citrus, grapes and apples), their potential for the generation of value-added products, as well as relevant research studies performed in South Africa, are summarised in Figures 1–4. A summary of potential beneficiation of agri-industrial wastes (solid and liquid) is provided in Figure 5.

South African studies on bio-based (fruit) wastes

Various studies have been carried out on wastes from the South African fruit industry to address aspects of waste treatment and beneficiation (Table 2). Work conducted on olive, citrus, grape and apple waste, is summarised in Figures 1–4. Many of these studies have taken place with funding obtained from the Water Research Commission of South Africa and the Wine Industry Network of Expertise and Technology (Winetech).

Burton et al.[72] carried out a feasibility study on the potential for energy generation from wastewater. They identified fruit industry wastewater as one of three wastewater sources with the greatest potential as sources of renewable energy. Fruit processing in South Africa includes canning, juicing, winemaking and fruit drying. Heavy water consumption occurs during these processes (7–10.7 m^3/tonne of raw produce) and the wastewater generated typically contains particulate organics, suspended solids, various cleaning solutions and softening or surface-active additives.[72] A compositional analysis of wastewater from an industrial fruit processor in the Western Cape Province revealed that fruit-processing wastewater could be a feasible feedstock for the production of bio-ethanol and biogas, but factors such as COD levels,

Fruit profile: Olives

Data on olive processing

South African 2012/2013 season: 2000 tonnes processed for olive oil production; 1500 tonnes processed for table olive production[11]

For olive oil production[14]: 3-phase – 80-120 L olive mill wastewater and 55 kg olive husk per 100 kg olives processed; 2-phase – 80 kg olive mill waste per 100 kg olives processed (one waste stream; solid and liquid combined)

Solid and liquid waste composition[14]

Press process: Solid and liquid waste

- Solid: fats and oils (~9%), proteins (~5%), total sugars (~1%), cellulose (~24%), lignin (14%), phenolic compounds (~1%)
- Liquid: low pH (4.5) and slightly more total sugars (~2.6%)

2-phase process: Solid waste only

- 56.8% moisture content compared to 27.2% from the press process; total sugars (~1%), cellulose (~14.5%), lignin (~8.5%), phenolic compounds (~2.43%)

3-phase process: Solid and liquid waste

- Solid: moisture content at 50.23%, total sugars (~1%), cellulose (~17%), lignin (~10%); phenolic compounds (~0.3%)
- Liquid: low pH (4.8) and 1.61% total sugars

Potential uses of olive-processing waste and potential for production of value-added products

- Bio-energy[15]
- Enzymes[16]
- Direct soil application (fertiliser/carbon sequestration)[17]
- Animal/fish feed[18,19]
- Composting[20]
- Pectin extraction[21]
- Source of polyphenols[22]
- Functional beverages[23]
- Sorbent for heavy metals from aqueous solutions[24]
- Novel materials, bio-plastics[25]

Research performed in South Africa

Focus of the studies: Bioremediation and beneficiation

Targeted waste stream: Table olive processing wastewater

Major conclusions/outcomes: Designed a bioreactor for the extraction of hydroxy-tyrosol, a potent antioxidant (beneficiation); microorganisms isolated from the waste showed great potential in the bioremediation of the wastewater.

References: 26, 27, 28

Figure 1: Olive waste profile – solid and liquid waste composition and potential for application and production of value-added products.

Fruit profile: Citrus

Data on citrus processing

South African 2011/2012 season[7]: Citrus (oranges, lemons, limes, grapefruit and naartjies) – 441 899 tonnes processed

Waste generated: > 1.5 ML of wastewater generated per tonne of citrus fruit processed[29]; Solid waste generated: peels, seeds, membranes and juice vesicles[30]

Solid and liquid waste composition

Liquid waste:

- 15% soluble solids and 30% pulp may be present; high COD (100-2000 mg/L) and biochemical oxygen demand (BOD; 20-1400 mg/L)[31]
- Wastewater nutrient concentration (in terms of phosphorous and nitrogen) is quite low, pH values are variable, various antioxidant compounds, as well as essential oils and heteropolysaccharides are present[29,31]
- The presence of terpenes in citrus-processing wastewater often makes biological treatment of the waste quite difficult because of the genotoxic effect of the terpenes on various prokaryotic and eukaryotic organisms[31]

Solid waste:

Citrus waste contains (g/100g)

- 8.1-10.1% total sugars
- 0.5-4.0% fat
- 7.0-12.5% protein
- 8.5-23% pectin
- 7.5-11.6% lignin
- 22.5-37.1% cellulose
- 5.60-11.0% hemicellulose[32]

Potential applications of citrus-processing waste and potential for production of value-added products

- Enzyme production[33]
- Biofuel production[34]
- Animal feed[35]
- Bioactive compounds such as antioxidants[36]
- Citric acid production[37]
- Heteropolysaccharide xanthum (gum)[38]
- Substrate for single cell protein production[39]
- Mushroom production[40]
- Composting[41]
- Ethylene production[42]
- Immobilisation carrier in solid state fermentation[43]
- Source of limonene and pectin[34]

Research performed in South Africa

Focus of the studies: Bioremediation and beneficiation

Targeted waste stream: Citrus solid waste and wastewater from citrus processing

Major conclusions/outcomes:

- Citrus solid waste can be used as composting material
- Organic oils present have potential application in cosmetics
- It is rich in antioxidants and antioxidant-rich dietary fibre
- Most of the waste streams are dilute, a pre-concentration step may be required and supplementation with solid waste would be required for full beneficiation

References: 13, 29, 41

COD, chemical oxygen demand

Figure 2: Citrus waste profile – composition of the waste and potential uses and application for the production of value-added products.

Fruit profile: Grapes

Data on grape processing

South African 2011/2012 season[7]: 1649 tonnes processed for preserves and canning; 151 628 tonnes are dried; 1 413 533 tonnes pressed for wine, spirits and juice production

Waste generated: 1 billion L wastewater generated per annum[44]; solid waste (typically 30% w/w of fresh grapes) includes skins and seeds (pomace or marc) and stalks[45]

Research performed in South Africa

Focus of the studies: Bioremediation

Targeted waste stream: Winery and distillery wastewater

Major conclusions/outcomes:

- Various treatment technologies can be applied for the treatment of winery wastewater
- Microorganisms and to a lesser extent, plants, play a key role in the treatment processes involving the use of constructed wetlands
- Upflow anaerobic sludge blanket reactors combined with ozonation, is a feasible treatment method for the bioremediation of winery wastewater
- Constructed wetlands (planted and unplanted) are inexpensive, low maintenance alternatives for the treatment of winery wastewater.

References: Various Winetech projects (1999-current)[46], including the following project numbers: WW 19/01, 120H, MV 04, US GMW, WW 19/05, WW 19/07, WW 19/08, WW 19/09, WW 19/11, WW 19/12, WW 19/13, WW 19/14, WW 19/15, CRSES 201227 and SU 2013/27; 44, 45, 46, 47

Solid and liquid waste composition

Liquid waste: Composition is variable and generally dilute; COD of 800-12 800 mg/L with glucose, fructose, ethanol and acetic acid contributing towards the COD[48,49]; low levels of organic acids and slowly biodegradable phenolic compounds; pH ~4.5.

Solid waste: Composition is variable, depending on the component and cultivar analysed; g/100 g: 1.8-3.7% soluble sugars; 1.8-3.8% protein; 0.3-1.0% lipids; 19.5-40.8% cell wall polysaccharides and lignin (dietary fibre)[45]

Potential applications of grape-processing waste and potential for production of value-added products

- Enzyme production[51]
- Biofuel production[52]
- Composting[53]
- Animal feed[54]
- Resin formulation[55]
- Pullulan production[56]
- Lactic acid and biosurfactant production[57]
- Substrate for growth of edible mushrooms[58]
- Source of polyphenolic and phenolic compounds (including antioxidants and pigments)[59, 60]

COD, chemical oxygen demand

Figure 3: Grape waste profile – waste composition and potential uses and application for the production of value-added products.

Fruit profile: Apples

Data on apple processing

South African 2011/2012 season[7]: 244 469 tonnes processed for juice, jams, preserves, etc.; 1110 tonnes dried.

Waste generated: Apple processing results in 25-30% solid mass or pomace and approximately 5-10% liquid sludge is produced[61]

Solid and liquid waste composition

Liquid waste:

The apple process sludge typically has a pH of 3.3±0.1, 115-135±5.0 g/L total solids, total nitrogen of 2.2-2.9 g/L, 44.3-51.9 g/L total carbon, 56.2-66 ±1.7 g/L total carbohydrates, 28.8-33.8 ±2.0 g/L protein, 5.1-5.9 g/L total lipids and various micronutrients[61]

Solid waste:

Biochemical composition of apple pomace is dependent on the variety of apple used as well as the stage of ripening at harvest[62]:

- 7.2-43.6% cellulose, 4.26-24.4% hemicellulose
- 15.3-23.5% lignin, 3.5-14.32% pectin
- 48.0-83.8% total carbohydrates
- 2.9-5.7% protein
- 1.2-3.9% lipids
- 10.8-15.0% total sugars (glucose, fructose and arabinose as major components and sucrose, galactose and xylose as minor components)[61]

Potential applications of apple-processing waste and potential for production of value-added products

- Enzyme production and induction[61]
- Biofuel production (ethanol and biohydrogen)[63]
- Incorporation into food products[64]
- Source of polyphenols (e.g. antioxidants)[63]
- Cultivation of edible mushrooms[65]
- Production of aroma compounds and pigments[63]
- Production of lactic acid[66]
- Production of citric acid[61]
- Substrate for microorganisms for the production of biopolymers[61]
- Animal/livestock feed[61]

Research performed in South Africa

Focus of the studies: Renewable energy, bioremediation and beneficiation

Targeted waste stream: Apple-processing wastewater, unprocessed fruit and apple pomace

Major conclusions/outcomes:

- Anaerobic ponds for the treatment of apple-processing wastewater are effective for the bioremediation of the wastewater and can be applied in the production of biogas (methane)
- Biogas production from excess, unprocessed fruit is feasible; due to seasonality of the waste, supplementation with another waste type was necessary
- Enzyme cocktails can be used for the breakdown of recalcitrant waste components; release of sugars sufficient for bio-ethanol production

References: 47, 67, 68, 69

Figure 4: Apple waste profile – composition of the pomace and sludge generated during apple processing, potential uses and applications in the production of value-added products.

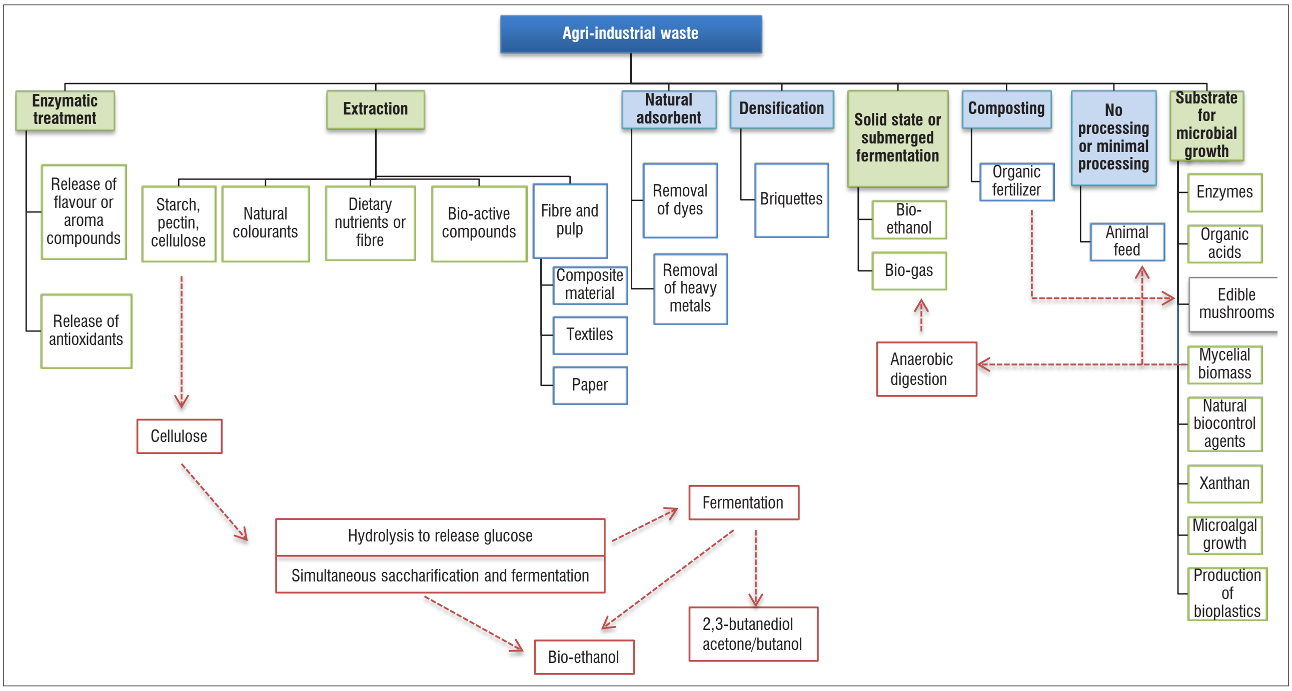

Figure 5: Potential beneficiation of agri-industrial wastes with a focus on fruit wastes. Green blocks indicate possible pathways for beneficiation of liquid waste and/or solid waste, while blue blocks represent possible beneficiation pathways for solid waste.

Table 2: Research performed in South Africa with a focus on bio-based waste (solid and liquid)

Focus of the study	Targeted fruit waste	Major conclusions/outcomes	Reference
Scoping study for bioremediation and beneficiation application	Pineapple cannery wastewater	The high carbohydrate content (19.8 g/L) of the wastewater makes this wastewater an attractive substrate for the production of yeast and/or ethanol.	73
Water and wastewater management in fruit- and vegetable-processing plants	General	A large volume of wastewater is generated during fruit processing; the report was written in the form of a guideline on how to minimise water intake and wastage.	74
Renewable energy	Fruit cannery wastewater	Biogas production from fruit cannery wastewater through the use of an upflow anaerobic sludge bed bioreactor is possible, but certain parameters need to be monitored (e.g. salt accumulation).	75
Renewable energy	Various fruit-processing wastes	Theoretical: Identified the fruit and beverage industries (brewery, distillery, winery, fruit juicing and canning) as one of three sectors for potential energy recovery from waste; sugar-rich wastewaters are a potential source for bio-energy production.	72

Note: Studies on olive, citrus, grape and apple waste are presented in Figures 1–4.
Sources: adapted from Dhillon et al.[61]*, Padam et al.*[70]*, Martin*[71].

sugar concentration and volumes generated should be considered during feasibility studies.[72]

An early study by Prior and Potgieter[73] explored the potential use of pineapple cannery wastewater and other fruit- and vegetable-processing waste as a substrate for the growth of a yeast strain and ethanol production. Pineapple cannery wastewater was found to be sufficiently high in carbohydrate content for bio-ethanol production. Binnie and Partners[74] mainly focused on water practices at fruit- and vegetable-processing plants. An evaluation of the water intake and wastewater generated, showed that excessive water wastage occurs in these processing plants. The study evaluated different types of processing plants receiving different types of fruit and/or vegetables, and presented a set of guidelines for these industries for the management of water intake and wastewater generation. However, industrial practices may have changed since these studies were undertaken and no new comprehensive evaluations have been performed since then. Subsequent studies on fruit waste (solid and/or liquid) have become more focused and aimed at bioremediation, beneficiation and/or renewable energy generation.

Interestingly, there is only one other reported study on the production of renewable energy from fruit cannery wastewater.[75] The main obstacle in the use of this wastewater stream is that the wastewater was found to be alkaline and contaminated with lye which is used during the canning process (for cleaning of tanks etc.). Sigge and Britz[75] were, however, successful in the application of an upflow anaerobic sludge bed reactor for biogas production but, over time, experienced excessive salt accumulation and ultimate failure of the bioreactor, indicating that an additional process for the removal of salts would be required.

On a larger scale, we are aware of one company in South Africa that utilises a fruit waste input. Brenn-O-Kem, with plants in Wolseley and Worcester in the Western Cape, successfully utilises grape pomace and lees from the wine production industry (grape processing) for the production of various valuable products.[76] These products include cream of tartar, calcium tartrate and grape seed extract. The remaining wastes after processing are dried and burned for fuel, which reduces the volume which is subsequently composted. Brenn-O-Kem is an excellent example of a company with a successful production strategy based on a sustainable waste stream.

Even though aspects of beneficiation and the application of fruit waste as a feedstock for renewable energy generation have been the focus of fruit waste studies in South Africa, there have been no studies regarding the production of enzymes as value-added products with fruit waste as the feedstock. The following sections will focus on the feasibility of fruit waste and waste streams as a feedstock for the production of value-added products, including industrially important enzymes.

Production of value-added products from bio-based (fruit) waste

Studies frequently cite production costs as one of the constraints in the scaling up of enzyme production for commercial or industrial exploitation. One of the most important cost considerations is the high price of culture media. In a recent study, Osma et al.[77] showed that for all the 46 different enzyme production systems they investigated, the cost of culture medium was consistently higher than the cost of equipment and the operating costs. It is therefore important for researchers to explore new and inexpensive media for enzyme production. Fruit waste streams can be potential substrates for the production of enzymes and other value-added products. Cellulase production, for example, may occur via solid-state fermentation or submerged fermentation.[78] Solid-state fermentation has many advantages over submerged fermentation as it requires less capital, lower energy, uses a less complex medium, results in higher productivity, requires less rigorous control of fermentation parameters, and produces less wastewater.[78] Krishna[79] made a direct comparison between solid-state and submerged fermentation using banana waste and found that cellulase production was 12-fold higher using solid-state fermentation. For reviews on solid-state fermentation see Couto and Sanroman[80] and Pandey et al.[81]

In many studies, the agri-industrial waste substrates were supplemented with further nutrients such as glucose, and/or nitrogen sources such as yeast extract or inorganic sources such as ammonium sulphate or sodium nitrate. Other supplements included mineral salts and trace elements. Supplementation with wheat bran is also common. The extent of supplementation is influenced by the substrate characteristics, as well as the growth requirements of the microorganism used. Where fruit-processing waste is used, the substrate may contain many of the minerals required, as well as residual sugars, and will therefore require less or no supplementation.[82] A viable alternative is the supplementation of the fruit-processing wastewater with solid waste to effect a bioremediation–beneficiation result.[13] Ideally there should be minimal supplementation in order to minimise costs.

Enzyme production through Aspergillus *strains on fruit wastes*

Local production of useful enzymes is encouraged under the new bio-economy strategy.[5] Currently, South Africa imports the majority of its enzyme requirements and the development of local manufacturing capabilities will decrease reliance on imports. Not only will this decrease the cost of enzymes, but the cost reduction will encourage their use in the development and establishment of environmentally sustainable industrial processes. Industrially important enzymes are a strategic area of interest as their use can translate to reductions in water usage, energy consumption, greenhouse gas emissions and other toxic waste emissions.

Aspergillus spp., notably *A. niger* and *A. oryzae*, have been used in the Orient for more than 2000 years for the production of fermented food and products such as citric acid and soya.[83] These fungi produce copious amounts of enzymes that can hydrolyse starch, pectin and cellulosics.[84,85] *Aspergillus* spp. can also degrade and utilise a wide range of phenolic compounds[86], including compounds present in olive mill wastewaters[87-89]. The ability of *Aspergillus* spp. to produce extracellular enzymes in large quantities and to utilise recalcitrant phenolic compounds, make them ideal for degrading more complex organic matter in waste streams.

Aspergillus niger has long been used for industrial enzyme production, in particular by companies such as Novozymes and DSM, and is the preferred organism for industrial enzyme production. Various *A. niger* strains capable of overexpressing cellulases, xylanases, mannanases[90,91] and a laccase[92] have been developed and tested. Enzyme production in grams per litre was demonstrated for a mannanase.[93] Furthermore, studies showed the production of cellulase and xylanase by *A. niger* strains cultured on the waste lignocellulosic streams remaining after fermentation of sugarcane bagasse and northern spruce.[94,95] In principle, it should be possible to grow *A. niger* strains on spent fruit waste streams after ethanolic fermentation, and on olive mill waste streams, with the simultaneous production of high-valued enzymes.

Conversion of agri-industrial wastewater to bioenergy

The first version of the Biofuels Industrial Strategy was released in 2007 with the overall aim of contributing up to 50% of the national renewable energy target of 10 000 GWh[96] through 4.5% blending of biofuels with petroleum. Before the release of the final strategy, commercial sugar producers and maize farmers represented the majority of the parties looking to drive the South African biofuels industry. However, the final Biofuels Industrial Strategy reduced the target to 2% of the liquid road transport fuels market. A 2% mandatory blending (for implementation from 1 October 2015) was only gazetted in 2012.

To date, the Department of Energy has issued and granted nine licences for the production of at least 500 million litres per annum of bioethanol and biodiesel from grain sorghum, soybean and waste vegetable oils. The biofuel plant to be built in Cradock in the Eastern Cape Province, funded by the Industrial Development Corporation of South Africa, has received the most attention as it will be seen as a case study for the nascent biofuels industry. In the first phase, 225 000 tonnes of grain sorghum will be imported from around the country and the second phase will use the produce from local farms, purchased by the Department of Rural Development and Land Reform. Mainly sugar beet and sorghum will be used to produce 90 million litres of bioethanol a year. The development of these biofuels facilities appears to be delayed by financing, availability of suitable land, incentives and policy decisions. Unfortunately, none of the initiatives of the initial stakeholders (maize and commercial sugar producers) has become established, mainly because of the Strategy's restrictions on the type and source of feedstock, as well as on the type of farmers (subsistence versus commercial) who would be subsidised. Considering the sensitivity of the food versus fuel debate, as well as sensitivity around land use and ownership, feedstocks outside these contentions would be ideal for biofuels production. The use of fruit waste for bioethanol production does not affect food security or land use, is readily available and, as a value-added by-product of wastewater treatment, is economically beneficial to industries.[97]

Potential production of biofuels from fruit waste streams

A variable portion of fruit waste contains fermentable sugars that can be directly converted to ethanol. In the case of fruit streams, the bulk of the fermentable sugars are hexoses that can readily be fermented with industrial strains of *Saccharomyces cerevisiae* (bakers' yeast). Several previous reports alluded to the potential use of sugar-rich fruit wastewater for the production of bioethanol but all concluded that the sugar content (typically $<10\%$) needs to be higher to ensure minimum ethanol levels of 4% to make distillation cost effective.[13,72,73] The major challenge would be to concentrate the wastewater streams to about 20% sugar, which is optimal for ethanol production.[98] If fruit streams could be handled or sorted such that high sugar streams are available, direct fermentation to ethanol could be one approach to produce ethanol for in-house energy generation, or for local use in ethanol-gel, a safe and renewable replacement for kerosene.[99] Examples of ethanol production from fruit wastewater have been reported from apple pomace[100] and citrus leachate[101]. However, because of the variable and inevitably low sugar concentration of fruit waste streams, enzyme hydrolysis is required to release more fermentable sugars from starch, pectin and cellulosics in the waste streams to boost sugar concentrations to levels of 20% and higher.[34,102-105]

Current status of advanced cellulosic ethanol technologies

Advanced technologies for the conversion of lignocellulosics to ethanol are slowly but surely coming to fruition. Several companies have demonstrated novel processes for the conversion of woody biomass to ethanol and the first commercial plants in the USA and European Union were commissioned in 2013.[106,107] A large variety of thermochemical and biochemical routes (as well as hybrids of both) are exploited by different companies, although production of bioethanol represents the largest portion of commercial initiatives with major players such as Abengoa Bioenergy, Beta Renewables, DuPont Biofuel Solutions and POET exploring simultaneous saccharification and fermentation processes using commercial enzymes and primarily herbaceous feedstocks, notably corn stover and cobs, switchgrass or *Arundo* reeds.[106] Mascoma, in partnership with Valero, is the only company exploring consolidated bioprocessing (one-step conversion of lignocellulosics to ethanol), using a proprietary recombinant yeast strain that produces key cellulase enzymes and which can utilise both hexoses and the pentose sugar xylose (called CBP yeast). Mascoma is focusing on hardwoods and pulps as feedstocks.[107]

In South Africa, there have been several breakthroughs in expressing cellulases in the yeast *S. cerevisiae*[108-113] and in the development of a consolidated bioprocessing yeast strain capable of converting pre-treated hardwood to ethanol with significantly reduced enzyme addition[114]. Pre-treatment of different agricultural residues, with the aid of a 15-L reactor steam gun, has been evaluated with South African sponsored research funding in anticipation of an emerging cellulosic ethanol industry.[115-118] Researchers have also developed the capacity to generate pyrolysis and gasification products from different cellulosic feedstocks to substitute fossil fuels such as coal, coking coal and reductants.[119,120] These studies also support developments toward the realisation of a bio-economy. Apart from the development of both biochemical and thermochemical technologies, expertise in process modelling, energy efficiency optimisation, economic viability assessment and life-cycle analysis have also been developed.[121] Such technology assessment is critical for both technology selection and technology integration into future biofuels/bioenergy/biorefinery industries.

Potential for biofuel production in South Africa

When advanced generation biofuels technologies come to fruition and 50% of the residual lignocellulosic biomass (almost 50 million tonnes produced annually in South Africa) can be used, biofuels could play a significant role in South Africa's transport fuel future. The potential contribution from different sources to the current total fossil fuel usage of 23 x 10^9 litres would be: 9.7% ethanol from agricultural residues, 3.2% ethanol from forestry residues, 10% ethanol from burned grasses (if these can be optimally utilised), and potentially 4.2% ethanol from the utilisation of invasive plants.[122,123]

The paradigm shift from fossil fuels to biofuels-generated energy will have far-reaching positive consequences; beyond the development of a sustainable energy resource, it will also impact on society. The decrease in levels of unemployment will play a major role in alleviating many social problems in South Africa related to unemployment and poverty.

A conceptual model to maximise utilisation of fruit-waste streams

The current management plans for many fruit wastes do not extract the full value from these wastes before disposal. In line with the bio-economy strategy, the full beneficiation potential of these wastes should be evaluated. Overall, the beneficiation potential of fruit wastes includes: extraction of valuable chemicals, provision of nutrient sources for the growth of alternative biomass (for either consumption or the production of valuable products like enzymes), feedstock for biofuels production, and composting or land application. These potential uses are not necessarily mutually exclusive and the full extraction value should be considered. Previous studies explored the utilisation of fruit and olive waste streams through different technologies. Although all the technologies have merit, none provides a complete solution in isolation. The concept of a biorefinery includes the separation of biomass resources (using a range of technologies) into their building blocks which can then be converted to a variety of value-added products. With this in mind, a non-exclusive biorefinery using fruit and olive wastes is proposed. This approach maximises the potential of fruit and olive waste streams through the integration of different technologies. This integration is shown as a conceptual model in Figure 6.

Combining fruit wastewater streams with lignocellulosic streams could overcome the limitation of both processes – that is, combining the low sugar content of fruit waste streams with the costly enzymatic conversion of lignocellulosics – in a manner analogous to the integration of ethanol

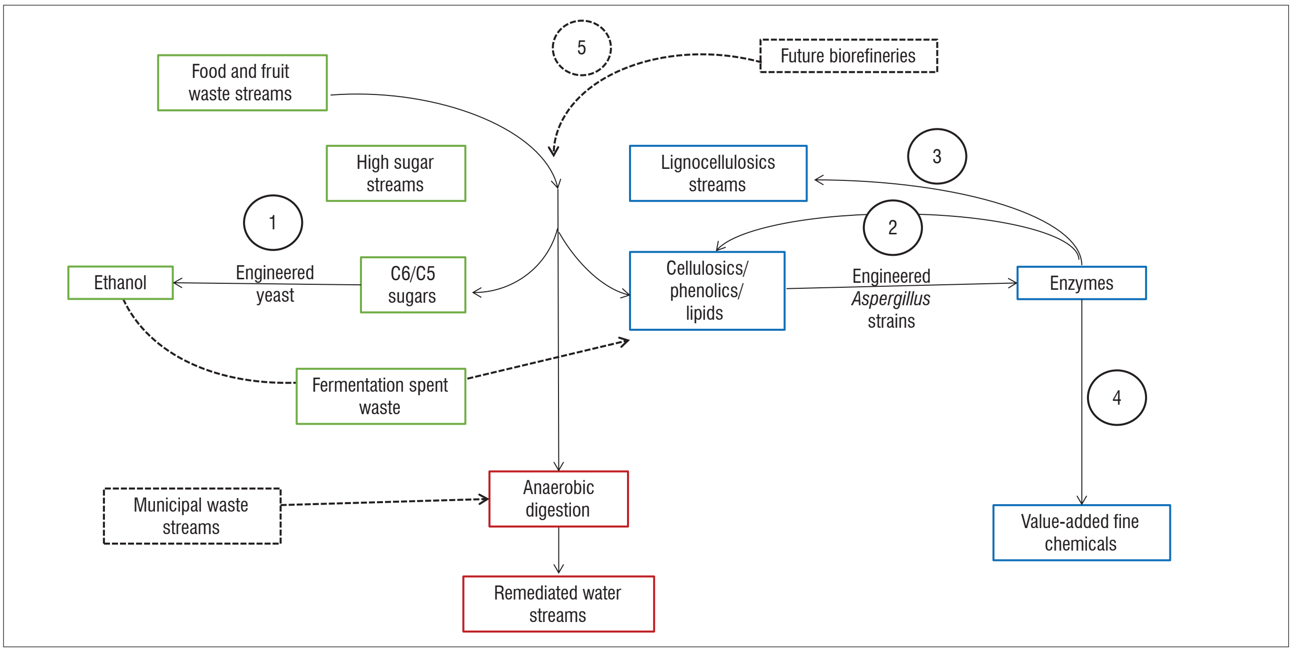

Figure 6: Integrated approach for remediation and beneficiation of fruit waste streams (ReBenFruWaste). Waste streams can be divided into (1) sugar-rich streams for ethanol production and (2) cellulosic/phenolics/lipid-rich streams for enzyme production by *Aspergillus* strains. Enzymes can be used for (3) bioconversion of lignocellulosic streams or (4) the production of value-added fine chemicals. The process can also include (5) biorefinery waste streams of future bio-economies.

production from sugarcane juice and sugarcane bagasse.[112] The ideal approach would be to first ferment high sugar streams to ethanol, or to combine such streams with lignocellulose sugar streams generated by employing commercial enzyme preparations, or enzymes produced with recombinant *Aspergillus* strains on spent fermentation streams. Subsequently, waste streams with lipids and high phenolic content (such as olive mill waste streams) or higher lignocellulosics (fruit slops or citric wastes) would be used to produce enzymes with recombinant *A. niger* strains. The remainder would be exploited for biogas production, or combined with municipal waste for biogas production.

Some disposal strategies for fruit waste (e.g. use as feedstock) utilise all components of the waste. Alternative beneficiation methods should also minimise waste in order to minimise environmental pollution. A biorefinery approach, with individual applications that may utilise different elements of the pomace, is capable of coupling complementary processes to achieve zero waste.

A fruit waste biorefinery should not only be able to produce valuable products, but also be sustainable in the long term and result in economic, environmental and social gains. The social and environmental impacts of some current disposal methods for most fruit wastes are clear: evaporation lagoons and direct soil application that lead to malodours and environmental toxicity if legal limits (which are often lacking) are not adhered to; ever-increasing transport costs for landfill disposal; as well as energy costs for drying wastes for animal feed. Aside from economic gains and decreases in environmental impacts, the implementation of beneficiation strategies in a fruit waste biorefinery could also result in social development in rural fruit-growing regions, with increased employment opportunities and skills development.

Disadvantages facing the application of the beneficiation technologies discussed are that fruit crops (and hence processing wastes) are seasonal, there are costs of transporting wastes for processing and there is variability in waste composition within and between fruit crops. These challenges can be overcome by centralising biorefineries in areas where fruit-processing plants are clustered and ensuring that the technologies used are robust and flexible enough to handle variable inputs. Centralising waste beneficiation plants in fruit-processing areas would reduce transport costs. For example, in South Africa, the grape and apple production and processing areas are in close proximity and a biorefinery that could extract the value from wastes from both crops would be advantageous. Furthermore, to decrease downtime, the processing of other agri-industrial wastes in the region could occur in the off-season/s.

Innovative solutions to overcome the challenges faced are required for the implementation and success of the new *Bio-economy Strategy.* Beneficiation of fruit wastes could play a role in the development of a bio-economy. It is important to note that the viability of waste source beneficiation is not determined by the ability to provide/fulfil all the needs of a country (for example, the entire biofuel requirements) in a particular sector; rather beneficiation should be considered feasible if the beneficiation potential is currently not being met and if a given process could contribute towards meeting these needs while resulting in decreased environmental impacts and positive social and economic gains.

Conclusion

In conclusion, it is clear that South Africa has vast resources in the form of fruit waste materials and waste streams that can be channelled into the production of various value-added products, notably biofuels and enzymes. The proposed conceptual model for an integrated system that utilises fruit and olive waste streams falls within the objectives of the South African bio-economy and the vision for the development of the industrial bio-economy and sustainable environmental management in South Africa (Figure 7). Not only does it address the extraction of bio-based chemicals and bio-energy, but also the need for bioremediation of (agri-) industrial wastewater. The conceptual model proposed (Figure 6) effectively addresses a number of strategic interventions laid out by the South African *Bio-Economy Strategy* which include the development of integrated biorefineries from bio-based feedstocks and strengthening of wastewater and solid waste research, development and innovation.[5] A successful integrated system would be beneficial for the country with regard to: (1) a reduction in the use of fossil fuels, (2) the treatment of waste streams and waste materials currently posing a threat to the environment, (3) job creation, (4) development of sustainable green processes and (5) the production of value-added products with the potential for South Africa to expand into a global, multimillion rand market.

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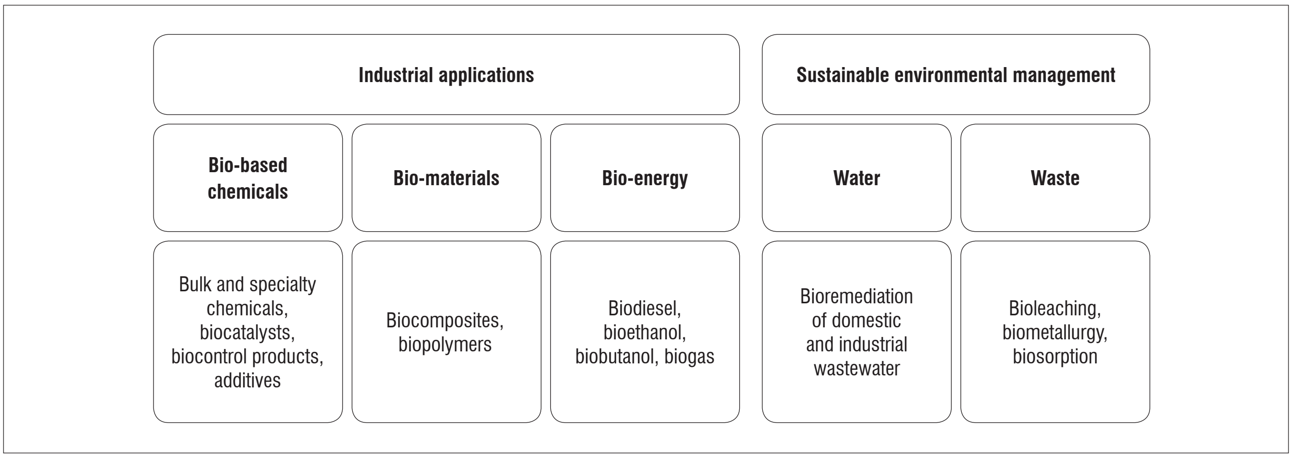
Authors' contributions

B.I.P was the project leader; W.H.v.Z. and M.L.R-H. were co-investigators on the project; and N.K., P.J.W., K.A.G., T.K., J.S.v.D and C.O. all contributed to the writing of the manuscript. Final proofreading and preparation of the manuscript was performed by N.K., M.L.R-H., W.H.v.Z and B.I.P.



Source: adapted from the Department of Science and Technology Bio-Economy Strategy[5]

Figure 7: Thematic areas that are the focus for the development of an industrial bio-economy in South Africa. Focus areas and products addressed by the conceptual model for the utilisation of fruit and olive wastes are highlighted in bold.

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