

JSAT

Journal of Science and Agricultural Technology

e-ISSN 2730-1532, ISSN 2730-1524

J. Sci. Agric. Technol.

Vol. 2 | No. 1 | January - June 2021

<https://www.tci-thaijo.org/index.php/JSAT>

Journal of Science and Agricultural Technology



J. Sci. Agri. Technol. (2021) Vol. 2 (1)

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Welcome message from Editor-in-Chief

Dear authors, reviewers, and readers

It is our great honor to present the first issue of the second volume of the Journal of Science and Agricultural Technology (JSAT), the official journal of the Faculty of Science and Agricultural Technology, Rajamangala University of Technology Lanna (RMUTL), Thailand. In this issue, the contribution consists of five articles from Australia, Indonesia, and Thailand. The JSAT has been published in Thai Journal Online (ThaiJO), indexed in Google Scholar, and Digital Object Identifier (DOI) under the National Research Council of Thailand. The journal will continue to publish high-quality articles under an intense peer-review process with supports from various educational institutions domestically and abroad. As an Editor-in-Chief, I promise to move forwards to gain international recognition preparing for a higher index ranking. Besides, I strongly encourage researchers to submit manuscripts to share knowledge and promote the growing field of science and agricultural technology. Lastly, I am so grateful for supports from our submitting authors, reviewers, and staff. Without you, the success of the current issue would not be possible.

Best regards



Assoc. Prof. Dr. Suntorn Wittayakun
Editor-in-Chief Journal of Science and Agricultural Technology
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ABOUT THE JOURNAL

Journal of Science and Agricultural Technology (JSAT) publishes original research contributions covering science and agricultural technology such as:

- Natural and applied sciences: biology, chemistry, computer science, physics, material science and related fields. Papers in mathematics and statistics are also welcomed, but should be of an applied nature rather than purely theoretical.
- Agricultural technology: plant science, animal science, aquatic science, food science, biotechnology, applied microbiology, agricultural machinery, agricultural engineering and related fields.

Furthermore, the JSAT journal aims to span the whole range of researches from local to global application.

The JSAT is published two issues a year.

Issue 1: January - June

Issue 2: July - December

Submissions are welcomed from international and Thai institutions. All submissions must be original research not previously published or simultaneously submitted for publication or submitted to other journals. Manuscripts are peer reviewed using the double-blinded review system by at least 2 reviewers before acceptance. There is no publication or processing fee.

The journal financial support is provided by Rajamangala University of Technology Lanna, Thailand.

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Agriculture: adapting to a changing climate

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Received: April 13, 2021. Revised: April 29, 2021. Accepted: May 3, 2021.

ABSTRACT

Climate change is having a severe effect on agriculture around the world. The seasons are shifting, droughts are increasing, and heavy rains and storms are intensifying. Australia, as a leading agricultural nation, is suffering severely from the impacts of climate change. In the past few decades, Australia has been devastated by prolonged droughts, damaging storms, forest fires, and severe flooding. Farmers, many of who were once sceptical, are now searching for answers. Yet agriculture is a significant contributor to climate change through anthropogenic greenhouse gas emissions and by converting non-agricultural land such as forests into agricultural land. Land cleared for agriculture and stock can no longer support or sustain the heavy stocking of sheep and cattle it once did. Crops are failing and heavy water use crops, such as cotton and rice, need to be reassessed or shifted to alternate areas. The immense irrigation areas where the bulk of Australian fruit is grown may no longer be sustainable. The tillage of soil and planting of broad-acre crops like barley, wheat, and oats, also needs to be managed differently in the future. If farmers are to remain viable in the next decade and beyond, traditional farming practices need to change, and farmers must find ways to mitigate the effects of climate change. While this may be problematic for some, it also opens exciting new ventures and infinite possibilities. This paper looks at the impact of climate change on Australian agriculture along with possible alternatives such as no-till sowing, hydroponic food production in the desert, and the use of red seaweed supplementation to ruminants in order to help mitigate the challenging years ahead.

Keywords: climate-change, mitigation, desert hydroponics, *Asparagopsis*

INTRODUCTION

Australia is a significant world agricultural leader with climatic regions ranging from tropical in the north to cold temperate zones in the south and everything in between. Although blessed with good natural resources and weather, Australia has always been a country of extremes. Recently, however, climate change has increased the risks. Drought is constantly present, as is extreme flooding. Cyclones have become more intense in the tropical regions, and in 2019-2020 Australia suffered the worst bushfires on record. Farmers have endured the burden of all these extremes, with many farms folding, breeding stock reduced or wiped out, and crops were failing. The agricultural sector, with its heavy reliance on water, is particularly vulnerable to climate variability and climate change. If Australia is to continue as a leading agricultural nation, we need to change our farming practices and strategies in order to adapt to a changing climate. Through a review of the available literature, this paper explores how farmers can better use current existing data, models, and information to better understand and adapt to agricultural processes to survive climate change. Two innovative solutions are investigated, including hydroponics in the desert

for large-scale tomato production and the use of Red seaweed (*Asparagopsis*) supplementation to reduce enteric methane in ruminants.

Climate change

Climate change may be the greatest challenge farmers will face over the next few decades. Before the Industrial Revolution started in the mid-1700s, the global average amount of atmospheric carbon dioxide was about 280 parts per million (ppm). The global average atmospheric carbon dioxide in 2019 was 409.8 ppm (Figure 1). Today, atmospheric carbon dioxide levels are higher than at any point in at least the past 800,000 years (Lindsay, 2020). This has mainly come from the increased burning of fossil fuels and industrialization, and extensive land clearing for grazing and other agricultural pursuits. Other greenhouse gases (GHG) such as methane are also problematic, with the livestock industry contributing up to 14.5% of GHG emissions (Gerber et al., 2013), with global methane emissions contributing to about 2.1 Gt CO₂ equivalent in 2010 (Smith et al., 2014). The result has been a rise in global temperatures and warming the planet with an increase of 1.41 degrees Celsius (IPCC, 2019). The speed of this change over such

a relatively short time period means evolution has not had the time to adapt to the changing climate and atmospheric conditions.

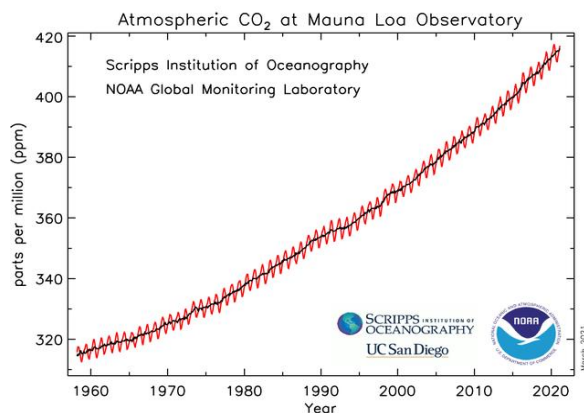


Figure 1. The global average atmospheric carbon dioxide
Source: NOAA (2021)

Drought in Australia

Australia has always been a country prone to drought. With a highly variable climate and low average rainfall, Australian agriculture is subject to more volatility than almost any other country in the world. There have been considerable changes to the Australian climate over the past 20 years, with reductions in average winter rainfalls in the south and increased temperatures (King et al., 2020). Climate models predict lower rainfall in southern Australia along with more severe droughts and floods in the future. (ABARES, 2021). Drought in Australia is commonplace, and it is one of the greatest challenges for any farmer resulting in severe crop failures and reduction in livestock feed. According to the Australian Bureau of Meteorology, drought events have increased and become more severe in recent years (BOM, 2020). With the land so dry, huge plumes of dust descended on the cities and towns, and the whole of Australia started to feel the effects of many long years of drought. Australia is in a difficult position in prioritizing climate adaptation in agriculture and investment because it is unclear how the IOD and El Niño–Southern Oscillation (ENSO) will change in the future and whether these changes will exacerbate drought conditions. Accurate projections of the future frequency of La Niña and negative Indian Ocean Dipole (IOD) events in a warming world will be required to better understand the risks of climate change on the security of Australia's water supplies, persistent droughts and extreme forest fires and how this will affect agriculture. These events may be increasing as the world warms (Zheng et al., 2013), but there remains

uncertainty in projections due to model deficiencies. In particular, climate models overstate the amplitude of the IOD (Weller and Cai, 2013) and struggle with the extent of La Niña (Taschetto et al., 2014).

Drought impacts on Australian agriculture in many ways. It reduces production in various agricultural sectors to well below levels experienced in non-drought years. Agricultural production impacts from drought include a reduction in farm income and an increase in farm debt. The drought of 1997 to 2009 caused a significant drop in income of up to 40% for both the grain industry and the beef industry (ABARE, 2004). During the same period, the dairy industry recorded the greatest loss of income in the 27 years that the statistics have been recorded (ABARE, 2005). Previous major droughts have seen income reductions of even greater proportions. Long-term investment losses of drought include removal of permanent plantations, orchards, and vineyards (Ejaz Qureshietal, 2013).

Fires

Climate change is already impacting fire seasons worldwide (Halofsky et al., 2020; Parente et al., 2018). As the number of dry and hot days increases, wildfire seasons are extending. A longer fire season is expected to result in more frequent and severe fires (Di Virgilio et. al., 2019; Matthews et al., 2012). There is emerging evidence from ecosystems worldwide that catastrophic events such as extreme drought and large bushfires can push terrestrial ecosystems past tipping points that result in abrupt ecosystem change (Davis et al., 2019). Given the impact of human-driven climate change on the frequency and intensity of these events there is a need to quantify their effects on plant and animal communities (Belzen et al., 2017) as they unfold. November 2019 was the driest month on record in Australia, and this was believed to be the catalyst for the horrific fires that followed in December 2019 and January 2020. During the summer of 2019-2020, the worst bushfires in recorded history spread throughout much of Australia. At least 18,983,588 hectares were burned, 3,113 houses lost, and 33 people died in the 15,344 bushfires that were collectively called the Black Summer fires. Millions of hectares of natural vegetation along the eastern coast, much of which had already been exposed to prolonged drought and recorded high temperatures, were burnt (Boer et al., 2020). Although it is impossible to provide a definite number, it is believed that over one billion animals died in the fires. On Kangaroo Island alone, around 100,000 sheep were killed by fires.

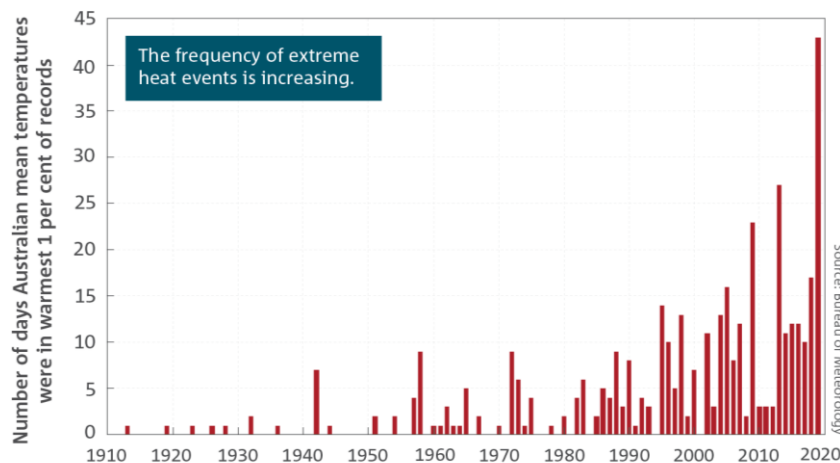


Figure 2. Number of days each year where the Australian area-averaged daily mean temperature for each month is extreme. Extreme daily mean temperatures are the warmest 1 per cent of days for each month, calculated for the period from 1910 to 2019
Source: BOM (2020).

How to mitigate the effects of climate change

Water

It is widely accepted that water is the most universally limiting factor in Australian agricultural production systems. Water efficiency, therefore, is one of the major initiatives in combatting climate change. In order to mitigate the problems farmers are facing due to climate change, they need to change the traditional methods of farming. No longer can Australian farmers rely on rivers and dams to irrigate their crops. Many traditionally irrigated farms may need to plant dry-land crops or use targeted trickle irrigation. The use of moisture probes before sowing and during the development of the crops has now become commonplace. No-till sowing has been used for many decades to prevent moisture loss from the soil, compared to traditional plowing, which exposes the soil to dry it out (Robinson, 2021). There has also been much discussion around some tree crops, such as almonds, which are heavy year-round water users. This industry has recently been criticized for its excessive water use to maintain the trees. Almonds use between 12 and 14 megalitres per hectare on mature orchards to produce 3.2 tonnes of the almond kernel and 6 tonnes of hull and shell that is predominantly used for cattle food. In a country where every drop of water counts, this may not be sustainable (Davies, 2019).

No-till cultivation

No-till farming is one cultivation method that has been shown to reduce soil erosion, maintain soil moisture and improve soil structure during the sowing of broad-acre crops and pastures. No-till farming, or conservation tillage, means that the land is not cultivated in the traditional manner prior to sowing. No-till cultivation helps farmers respond to

climate change by building up organic material in the soil, reducing water evaporation and runoff, and increasing soil carbon sequestration (Bayer et al., 2006). Other advantages include reduced tractor runs during sowing resulting in a saving on fuel, reduced wear and tear on machinery, and less carbon being emitted (Huggins and Reganold, 2008). For farmers in southern Australia, no-till's ability to help mitigate climate change while also adapting to the drier conditions makes it particularly relevant (Ugalde et al., 2007). The conversion to no-till farming systems in Australia has been both recent and rapid, with around 80-90% of Australia's winter broad-acre cropping farmers using no-till conservation methods (Bellotti and Rochecoste, 2014). The main reason there has been such a large uptake is mostly in response to soil erosion from wind and water (D'Emden et al., 2008).

However, although there are many advantages, there are also some disadvantages. Firstly, the changeover to no-till equipment can be expensive. There is also the issue of the use of herbicides to manage weed control (D'Emden et al., 2008). Many farmers are concerned that the herbicides will become less effective over time. With some herbicide companies controlling seeds, they may be limited in the use of other seed varieties (McRobert et al., 2011). One of the purposes of conventional tilling is to remove weeds, and no-till farming changes weed composition. In Australia, this is usually solved with the use of herbicides such as glyphosate instead of tillage for seedbed preparation. Weeds can also be controlled through winter cover crops, soil solarisation, or burning. Cover crops are sometimes used to help control weeds and increase soil residue. Extra fertilizers may need to be added due to the reduced mobility of nitrogen in the soil

(Bellotti and Rochecouste, 2014). Where herbicides are a problem, such as in organic farms, cover crops can be used. However, cover crops need to be killed to reduce competition and are usually done using rollers, crimpers, or other forms of weed control (D'Emden et al., 2008). Another difficulty can be residue from the previous year's crops lying on the surface of the paddock. This can cause different, more significant, or more frequent disease or weed problems than tillage farming. While the process of no-till cultivation is many thousand years old, the use of technology and innovation has made the practice very competitive, especially for broad-acre cropping, compared to traditional plowing and cultivation (Bellotti and Rochecouste, 2014). The process of innovation in farming is constantly changing, especially with the use of computerized soil management systems. New technologies such as Global Satellite Systems (GPS) and drones have been applied in ways not envisaged at the time of their invention. This type of farmer innovation continues to lead, using new applications and technology in the rapidly evolving domain of agriculture.

Hydroponics in the desert

One solution to the water issue may be using land previously thought unsuitable for farming and use the heat of the sun to help produce fresh water from normally unusable seawater. This is what is happening in the Australian desert. An enormous hydroponic farm has been established in the South Australian desert and now supplies up to 15% of Australian truss tomatoes. Sundrop Farm near Port Augusta in South Australia is the first of its type in the world and started with a small pilot plant in 2012. Now it is fully operational and uses solar energy to desalinate seawater and heat or cool the greenhouses to grow vine-ripened truss tomatoes commercially.

Seawater is piped 5.5 kilometers from the Spencer Gulf to Sundrop Farm in the arid Port Augusta region. A solar-powered desalination plant removes the salt, creating enough fresh water to irrigate 180,000 tomato plants inside the greenhouse (Klein, 2016). The farm uses 24,000 motorized parabolic mirrors arrayed at its base to project the sun's rays to a tower 127m above the ground. The thermal energy produced from this solar plant is used to heat seawater in vast boilers. This generates electricity from the resulting steam and thermal heating for the greenhouses. The steam-generated power also drives a desalination plant, turning constantly circulating seawater from the nearby Spencer Gulf into freshwater. In the greenhouses, 750,000 tomato plants are hydroponically grown in nutrient-filled pipes. The thermal energy harnessed here powers 20ha of adjoining greenhouses, which in

turn produce over 350 tonnes of tomatoes each week. The entire system is self-sustainable (Neals, 2016).

The Sundrop System uses the sun's energy to produce freshwater for irrigation in a closed-loop system. It is then turned into electricity to power the greenhouse to heat and cool the crops. The salt from the desalination plant is later sold as a by-product. Sundrop produces high-quality truss tomatoes that are distributed across Australia through a major supermarket chain. The beauty of this system is that it uses land and seawater, previously unusable, into a viable form of sustainable farming, conserving water, and totally free of fossil fuels. With Port Augusta having 320 sunlight days per year, there is no shortage of sunlight to power the system. One of the major costs is the cooling of the greenhouses during the summer. Although the initial cost of \$200 million is an extremely high capital investment, it is envisaged that the dramatic savings in running costs will make the investment work. Another facility is planned for Australia as well as facilities in Portugal and the USA in the near future.

Ruminants and enteric methane

Livestock production, particularly that of ruminants, is a large contributor to greenhouse gas emissions (GHG), particularly in the form of enteric methane. A review of mitigation options for enteric methane produced from ruminants showed that some of the effective strategies include increasing forage digestibility, replacing grass silage with corn silage, feeding legumes, adding dietary lipids and concentrates (Hristov et al., 2013). Another method being investigated is the use of red seaweed *Asparagopsis* as a feed additive. The seaweed from the genus *Asparagopsis* is a potent agent that reduces methane production in the digestive process of cattle and sheep. In general, marine algae were found to be more effective than freshwater algae in reducing methane production. Freshwater macroalgae have a similar biochemical composition to decorticated cottonseed meal (DCS); however, the methane output relative to DCS was reduced to 4.4% for *Spirogyra* and 30.3% for *Oedogonium* after 72 h incubation. However, there is no correlation between the biochemical composition of freshwater and a reduction in methane. Although methane was reduced, there were no apparent negative effects on fermentation variables. Rather, freshwater macroalgae had a slightly higher total volatile fatty acid (VFA) concentration than DCS with similar organic matter degradability (OMD) demonstrating that fermentation processes had not been compromised (Getachew et al., 1998). The effectiveness of the seaweeds in reducing enteric methane has now been established. However, only

Asparagopsis demonstrated that it remained effective and dramatically reduced the emission of methane, without negative impacts on rumen function. This was found even with a relatively low inclusion level in animal diets (Kinley et al., 2016; Li et al., 2018).

Asparagopsis taxiformis is a species of red algae with distribution in tropical to warm temperate waters. Native to the Southern hemisphere, it has been introduced to the northeastern Atlantic Ocean and the Mediterranean Sea. It is found widely in Australian waters, particularly around Northern Australia to Rottnest Island, Western Australia, and southern Queensland; Lord Howe Island; The Gulf region of South Australia, and Tasmania (Guiry and Guiry, 2021). *Asparagopsis armata* has also been used with good effects in cattle feed trials (Roque et al., 2019). The *Asparagopsis* species of seaweed produces a bioactive compound called bromoform, which prevents the formation of methane (CH₄) by inhibiting a specific enzyme in the gut during the digestion of feed. Supplements added to feed have been found to reduce enteric methane production by more than 80 percent in some animals (CSIRO, 2020). Roque et al. (2019) found 'there was no significant body weight change between cows receiving *Asparagopsis armata* at low inclusion compared to control; however, cows receiving the 1% level gained 9.72 kg less than control cows. Milk yield did not differ significantly between cows in the control group and those at a low level of *Asparagopsis* inclusion. However, cows fed at the higher level of *Asparagopsis* inclusion produced 11.6% less milk compared to control (P < 0.001). No significant differences were found in milk fat, lactose, solids non-fat, milk urea nitrogen, or somatic cell count with both levels of macroalgae inclusion. The conclusion of this study confirms that enteric methane emissions could potentially be halved by using seaweed as a feed additive to dairy cattle. (Roque et al., 2019). *Asparagopsis* seaweed is characterized by secondary metabolites with antibacterial properties and demonstrates a potent methane reduction effect in livestock digestive fermentation. Using low volumes (less than 1.0%) in a feedlot trial, methane was reduced by over 90%, with positive trends observed for feed conversion and productivity. The *Asparagopsis* species of seaweed produces special substances containing naturally occurring bromine (CHBr₃) that prevents the completion of methane construction by reacting with vitamin B₁₂ at the last step, which disrupts the enzymes used by the specific gut microbes that produce high energy methane gas as waste during digestion (CSIRO, 2020). The numerous studies on *Asparagopsis* as a feed additive to reduce methane

levels in livestock have now gone beyond the experimental phase and are being developed as a commercial product by the Commonwealth Scientific and Industrial Research Organisation (CSIRO) in Australia (CSIRO, 2020). Seaweed production globally is increasing, with more than 25 million tonnes (measured when wet) farmed each year. Australia currently has almost 1 million feedlots and 1.5 million dairy cattle. In order to produce enough *Asparagopsis* seaweed to supplement the feed of just 30% of these cattle would require about 25,000 dry tonnes a year and hundreds of thousands of tonnes if it were to be scaled up globally. With the selection and breeding of seaweed varieties for higher bioactivity, this figure is likely to reduce, but perhaps only by half, and it would still require large areas of land and water. With typical seaweed production rates at 30-50 tonnes of dry matter per hectare, this suggests that to supply 30% of the Australian feedlot and dairy industry will require approximately 2,000 hectares of seaweed farms. Seaweed farms in Australia are likely to be part of our increasing demands on the marine environment and will need to be part of integrated ecosystem-wide management and marine spatial planning. Indirect benefits worldwide, include creating alternative livelihoods in developing countries where fishing may be in decline, an alternative enterprise for existing aquaculture operations, and the use of seaweed as a means to filter detrimental nutrients from rivers or effluent from fish farms. In 2020, The Australian Seaweed Institute released its Blueprint for Growth, listing the cultivation of *Asparagopsis* as a key opportunity (Kelly, 2020).

CONCLUSIONS

The aim of this paper is to provide an overview of the effect climate change is having on Australian agriculture, and how through innovation, solutions can be found. Two examples are used to show how originality can be used to overcome seemingly insurmountable problems. Freshwater is a scarce commodity of which many countries do not have enough for drinking water, let alone irrigation for crops. By using the power of the sun to convert abundant seawater into freshwater, not only can this water be used in greenhouses in the desert to produce fruit and vegetables, but other uses, such as desalination plants, could be found for the systems in arid regions. The continuous rise of GHG, including the large contribution from livestock products, has long concerned scientists. The novel use of seaweed supplementation in trials has found that it can decrease the methane produced by ruminants. This will be of great assistance to livestock producers

worldwide. Additionally, new opportunities will be developed for seaweed farmers in Australia and worldwide to grow enough supplements to feed the world's cattle. Furthermore, the growing of the seaweed will have additional benefits in lowering GHG from the carbon dioxide used during the growing phase.

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Soybean (*Glycine max* L.) as a vitamin rich food to boost immune system for post-pandemic era

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Received: April 2, 2021. Revised May 16, 2021. Accepted: May 17, 2021.

ABSTRACT

The immune system of the human body plays a significant role in preventing COVID-19. Gibson et al. (2012) stated that the consumption of vitamin-rich and functional foods could boost the immune system to suppress the virus, especially in this pandemic era. This pandemic also affects the agricultural sector in Indonesia, especially in food security, supply, and chain. Coping with this problem, the Indonesian government has introduced *Gerakan Ketahanan Pangan* (GKP) program to support the agricultural sector. The program focuses on primary and functional food, particularly soybeans (*Glycine max* L.). The nutrition in soybeans may become the substitute for red meat, fish, and egg that considered expensive for several people. This situation may enhance soybean farmers, specifically in Indonesia, to face the post-pandemic period anticipating the return of similar viruses or conditions. This study was a qualitative analysis using a constructivist paradigm, and the data used were secondary data from literature and narrative review. The results of this study highlighted the need for immune-boosting food, especially soybeans. Shortly, soybeans are considered a long-term commodity that helps the food security and improves the Indonesian economy during the COVID-19 pandemic situation.

Keywords: immune system, food security, pandemic, soybeans, agriculture

INTRODUCTION

COVID-19 has claimed more than 500 billion souls worldwide, and the spread of COVID-19 has increased rapidly. COVID-19 attacks the human respiratory system. This situation impacts human health and leads to a global food crisis (Balkan, 2020). With the discovery of a new kind of Coronavirus variant, researchers are required to make vaccines that took a long time. Researchers have attempted to improve the immune system against this COVID-19. Especially, humans can boost their immunity with natural food crops. Several actions and policies have been implemented to address food security (Ge et al., 2018; Blay-Palmer et al., 2013; Johnson et al., 2012). Affordability, availability, and food accessibility for the communities have focused on the government, the private sector, and other stakeholders (Deng et al., 2015).

On the other hand, consumers try to protect themselves and improve their immune systems and are concerned about changing their food consumption habits, bioactive ingredients, and nutritional content (Galanakis, 2020; Rizou et al., 2020; Zinoviadou et al., 2015). However, security protocols are required applied following health standards for the prevention of COVID-19 (Rizou et al., 2020). One of the industrial sectors that had to overcome different

challenges during the pandemic is the food sector, striving to produce and secure sufficient and safe food. Food security, food safety, and food sustainability are recognized as strongly affected dimensions of food systems during the COVID-19 pandemic (Galanakis, 2020). Finally, food ingredients and bioactive compounds supporting immune functions in humans, such as vitamin C, vitamin D, polyphenols, and flavonoids have also been in research focus in preventing and treating COVID-19 as outlined in the review by Djekic et al. (2021). A multi-country study in 16 countries identified health as one of six determinants in eating behaviors (Djekic et al., 2021), so scientific evidence of the promising effects of food supplements and nutraceuticals can help citizens in protecting themselves during the pandemic and post-COVID-19 pandemic era.

Therefore, to protect humans during the pandemic and prepare for the post-pandemic era, consuming food with ingredients supporting the immune system is essential, such as soybeans. Soybeans represent a remarkable source of high-quality protein, vitamins, and minerals with a low saturated fat content and high in dietary fiber. Soybean protein is considered a suitable substituent for animal protein. Except for sulfur amino acids (methionine and cysteine), their nutritional profile is

almost similar to animal protein because soybean proteins contain most of the essential amino acids for animal and human nutrition (Hassan, 2013). Soybeans also contain a high level of isoflavone and vitamin C as an immune booster for the human body. This statement is supported by Makmun and Rusli (2020), who state that vitamin C or known as ascorbic acid, acts as an antidote for free radicals (pathogens) that could enter the body like the COVID-19 virus. A high concentration of isoflavone in soybeans is beneficial for health, such as anti-inflammation, anti-cancer, anti-allergic, antiviral, preventing osteoporosis, and lower heart disease risk, and could block the potentially harmful effects against the excess estrogen production in the human body (Krisnawati, 2017).

Furthermore, the agri-food chain is labor-intensive and can be affected by limitations in the workforce due to infection and/or limited ability to travel (Delhey and Dragolov, 2014). There should be efforts to allow for global trade and overall to minimize logistics disruptions so that major staple commodities can be moved across countries. Availability of food products is expected to affect the nutritional habits of consumers directly. It would be worth ensuring that diets during this crisis provide the necessary macro and micronutrients.

MATERIALS AND METHODS

The research approach we use is the qualitative approach with a descriptive method to analyze the literature review research. Descriptive research is a method that is intended for describing phenomena that have occurred recently or happened in the past. According to Furchan (2004), descriptive research tends to represent a phenomenon's existence by reviewing regularly, prioritizing objectivity, and doing carefully. Several aspects of the focus of the information collected in this study include the role of soybeans in health aspects, production and financial effects, food security, and the influence of soybeans on the body's immune system. The scope of the research location as a reference and case study data collection used is the Asian area, especially Indonesia.

RESULTS AND DISCUSSION

Soybeans are one of the world's most important crops with multifunction uses, including food, feed, fuel, and other industrial usages such as paint, ink, and plastics. Soybeans themselves also have various roles in our livelihoods, including in health aspects, production and financial effects, food

security, and the influence of soybeans on the body's immune system.

The role of soybeans in health aspects

Soybeans are used for thousands of years ago, especially in the food industry. Currently, the global soybean production is up to 219.8 million metric tons (Gandhi, 2009). The values of soybean in human nutrition and health are used in human foods in various forms, including infant formulas, flours, protein isolates and concentrates, and textured fibers. New soy foods are continually developed. Consumption of soy foods is increasing because of reported beneficial effects on nutrition and health. These effects include lowering plasma cholesterol, prevention of cancer, diabetes, and obesity, and protection against bowel and kidney disease. Soy-based infants are widely used to feed children suffering from allergies to cow's milk and prevent illness when breast milk is not available. The soy formulas are inexpensive and nutritionally adequate to replace milk-based formula and rarely elicit allergic reactions.

The role of soybeans in production and financial aspects

According to Chianu et al. (2010) many countries import soybeans as it became the number one priority of protein source and the second-highest for its oil (vegetable oil). Exporter countries import soybean, whether it's in the form of grain, biodiesel, or even soybean production, to another country with high demand. In 2001-2003, the world supply/demand for soybeans reached 183.9 million tons. About 10% of them were directly consumed as food (5.9%) and for feed (3.8%), and 84.2% were processed into soy oil and soy meal. Despite the fact that soybeans are mainly used as an industrial product in the form of feed, fiber, fuel, and food ingredients, they can be found in Asian and specialty markets as final consumer goods (Masuda and Peter, 2009).

According to Voora et al. (2020), soybean stocks maybe even higher than predicted, given the effect of COVID-19 on the food supply chain. This might affect the soybean prices if accumulated soybean stocks of soybean get dumped all at once into international markets. Furthermore, since the soybean market can be segmented into animal feed, food and beverage, personal care, dietary supplements, pharmaceuticals, and biomaterials, including biofuels, the versatility of soybeans will be a major factor in maintaining demand growth. From the information above, soybean will become one of the most potent food markets and also a higher

demand as the population keeps growing in the future (Wang, 2017).

The role of soybeans in food security aspects

In Indonesia, the government has launched a Food Security Movement program during the pandemic. Through the Agricultural Human Resources Extension and Development Agency, the Ministry of Agriculture has formulated 4 Ways of Action Method to achieve food security. First, increasing production capacity. The Ministry of Agriculture invites agricultural actors to accelerate rice planting in Season II 2020, covering an area of 6.1 million ha, development of swamps in Central Kalimantan Province 164,598 ha, including the intensification of 85,456 ha of lawn and extensification of agricultural land 79,142 ha. Second, diversification of local food. The Ministry of Agriculture will develop local food diversification based on local wisdom that focuses on one main commodity. Third, strengthening food reserves and logistics systems by provincial government rice reserves (CBPP), then strengthening regency/city government rice reserves (CBPK). Fourth, the development of modern agriculture, through the development of smart farming, are developing the use of screen houses to increase the production of horticultural commodities outside the planting season, the development of farmer corporations, and the development of food estates to increase the production of main food (rice/corn) (Caron and Kalafatic, 2016).

Immune link with COVID-19

The immune system becomes vital once an individual is exposed to an infectious agent. However, the nature of contagious agents varies, and the immune system requires different approaches to deal with different types of infectious agents. These different approaches follow similar general strategies, which aim to seek out and destroy, but the precise immune mechanisms involved can differ. Thus, the roles for nutrients in supporting the function of the immune system are many and varied, and it is easy to appreciate that an adequate and balanced supply of these is essential if an appropriate immune response is to be mounted. Nutrition contents in soybeans can be seen in Table 1.

Table 1. Nutrition contents in soybean seeds per 100 g.

Nutrients	Amount
Energy	1,866.0 kJ (446 kcal)
Carbohydrate	30.16 g
Sugar	7.33 g
Fiber	9.3 g
Fat	19.94 g
Monounsaturated	4.404 g
Polyunsaturated	11.255 g
Protein	36.49 g
Tryptophan	0.591 g
Threonine	1.766 g
Isoleucine	1.971 g
Leucine	3.309 g
Lysine	2.706 g
Methionine	0.547 g
Phenylalanine	2.122 g
Tyrosine	1.539 g
Valine	2.029 g
Arginine	3.153 g
Histidine	1.097 g
Alanine	1.915 g
Aspartic acid	5.112 g
Glutamic acid	7.874 g
Glycine	1.880 g
Proline	2.379 g
Serine	2.357 g
Water	8.54 g
Vitamin A	1 mg
Vitamin B6	0.377 mg
Vitamin C	6.0 mg
Vitamin K	47 mg
Calcium	277 mg
Phosphorus	704 mg
Potassium	1797 mg
Magnesium	280 mg
Sodium	2 mg
Iron	15.70 mg
Zinc	4.89 mg

Source: Wardani and Sujana (2020)

In essence, good nutrition creates an environment in which the immune system can respond appropriately to challenges, irrespective of the nature of the challenge. Conversely, poor nutrition creates an environment in which the immune system cannot respond well. The innate immune system acts fast (in minutes) after it recognizes a pathogen and, in most cases, eradicates

the invading pathogens. During this process, the cells of the innate immune system, and their derived immune mediators/proteins, also activate the cells of the adaptive immune system which then develop memory immune responses toward these pathogens. Therefore, upon reinfection, the intensity of the innate immune system remains the same.

Food is one of the factors that can affect the immune system. The three main categories of food that can boost the immune system, one of those is vegetables, berries, and nuts. These foods are categorized as functional food (Smith and Charter, 2010). As previously known, soybeans are a food or food ingredient that contains lots of vitamins. The vitamins contained in soybean include water-soluble vitamins, such as thiamine, riboflavin, niacin, vitamin C, pyridoxine, biotin, and folic acid. Second is fat-soluble vitamins, such as vitamin A, E, D, and K (Hassan, 2013; François et al., 2020). These vitamins have benefits as an immune booster because of their respective functions and reactions to the human body.

Immuno-nutrition plays an essential role in regulating either the action of the immune system or the activation effects of the immune system on specific nutrients or foods. (François et al., 2020). It is known that undernourished people have low immune functions to provide a guard against pathogenic species (Derbyshire and Delange, 2020). There are no medicines or nutrients capable of optimizing the immune system. However, there are methods, supported by scientific evidence, that can strengthen our immune systems, such as exercise, diet or nutrition, and immunity, and mental wellbeing and immunity, which result in better health and better quality of life, as described above. (Calabrese and Fasenmyer, 2017).

Soybean vitamins role as an immune booster

Soy vitamins are micronutrients that play an important role as cofactors in the metabolic machines involved in the generation of energy and biosynthesis required for immune response and multiple micronutrient supplementations immune-supporting functions, which can modulate immune function and reduce the risk of infection. (Gibson et al., 2012; François, et al., 2020; Gombart et al., 2020). It can be inferred that micronutrients play an important role in the need to defend or protect against infections such as COVID-19. Soy vitamins such as A, C, K, and B6 may play a role in preventing this pandemic. About micronutrients in soybeans can be seen in Table 2.

Fauziah et al. (2016) researched soybeans using the visible spectrophotometry method to determine the vitamin B1 content in soybean; in determining the wavelength maximum, vitamin B1

absorbance obtained a maximum of 0.678 in length wave 423 nm. Once obtained, the maximum absorption wavelength then measured the absorbance of vitamin B1 on concentration 1.25 ml (10 ppm), 1,875 ml (15 ppm), 2.5 ml (20 ppm), 3.125 ml (25 ppm), 3.75 ml (30 ppm). From this measurement gets the equation of the line straight i.e. $Y = -0.0020 + 0.0228X$ and the correlation coefficient (r) is 0.9997. Since the value of the correlation coefficient ($r \leq 1$), then the calibration curve obtained is linear. From this equation, the level of the total vitamin B1 can be determined from the sample solution.

The several literature reviews of nutrient analysis in soybeans show that soybeans contain many beneficial vitamins. These vitamins are B1, C, E, and B12. Vitamin B1 content based on the study research can be found by visible spectrophotometry. From the measurement of the calibration curve, obtained a standard deviation (SD) of 0.0066 limits of detection (BD) 0.8684 µg/mL and limit quantization (BK) 2.8947 µg/mL. Accuracy is measured as a coefficient variation (CV).

Table 2. Nutrition contents in soybean seeds per 100 g.

Items	Nutrient value ¹
Water soluble vitamin ²	
Thiamin (B1)	0.874 mg
Riboflavin (B2)	0.870 mg
Niacin (B3)	1.623 mg
Pantothenic acid (B5)	0.793 mg
Pyridoxine (B6)	0.377 mg
Folic acid (B9)	375 µg
Fat soluble vitamin ²	
Vitamin A	22 IU
Vitamin C	6 mg
Vitamin E	0.85 mg
Vitamin K	47 µg

Source: Haytowitz et al. (2019)¹; Saghir et al. (2017)²

Soybean fat-soluble vitamins, vitamin A, have a significant effect on human immune response, deficiency of vitamin A can change in immune response, increased susceptibility to disease or infection, and loss of membrane function in the immune system (François et al., 2020). Vitamin D also contributes to the innate of adaptive immune responses as it enhances phagocytosis, superoxide output, and bacterial killing of innate immune cells. This vitamin will play a role in the prevention and recovery of COVID-19 because, if we have lower amounts of this vitamin in the body, it has been shown to cause a rise in infection rates and has documented to increase the risk of coronavirus

infection (François et al., 2020; Nonnecke et al., 2014; Azrielant and Shoenfeld, 2017; Aslam et al., 2017). The role of vitamin E in the immune system is the vitamin needed to prevent the high risk of oxidative damage to immune cells against oxygenizing agents and to improve their physiological activity, and reduce immunosuppressive factors such as PGE2 prostaglandin (Meydani et al., 2005; Pae et al., 2012). Vitamin E supplementation may modulate host defense against infectious pathogens like COVID-19 (Meydani et al., 2004).

Water-soluble vitamins in soybeans, such as vitamin B6 (pyridoxine), folic acid, and vitamin B12 (cobalamin), which are only used in fermented soya products, including tempeh, are supported by natural killer cells in the immune system, making them essential for antiviral protection. The body requires B6 to consume vitamin B12 and to make the red blood cells and cells of the immune system (Aslam et al., 2017). Vitamin B6 helps to boost the immune response to increased antibody synthesis and also helps in communicative interactions with cytokines and chemokines (Kunisawa and Kiyono, 2013). Vitamin B12 (cobalamin), which is present only in fermented soy products, acts as a destroyer or affects pathogens activity to create infection by enhancing the work of B-cells and T-cells in the immune system (François, et al., 2020; Rowley and Kendall, 2019). As a result, because of its potential to influence the pathogen process that causes infection, cobalamin can allow COVID-19 patients not to be easily threatened by the virus. Vitamin B9 (folic acid or folate) is a vitamin that can boost the body's ability to fight COVID-19 due to its ability to sustain or enhance NK cell cytotoxic activity and its involvement in the intestinal immune system (François et al., 2020; Gombart et al., 2020).

Other vitamins, thiamine (vitamin B1), are used to improve the immune system. Low levels of vitamin B1 (thiamine) in our body can contribute to a lack of immune response in patients with diabetic and chronic tonsillitis (Mahmodi and Rezaei, 2019; Dakshinamurti, 2015; Aleszczyk et al., 2001). Vitamin B2 (riboflavin) maintains healthy blood cells and boosts metabolism. This vitamin is an efficient anti-inflammatory modulator, stimulates phagocytic macrophage function, and its metabolites are involved in MAIT cell stimulation (Mahmodi and Rezaei, 2019). Therefore, the importance of riboflavin in health and disease is becoming apparent and can help prevent COVID-19 from occurring in the human body. Vitamin B3 (niacin) is widely prescribed for its anti-inflammatory role in inflammatory disorders. Due to the existence of

nicotinic acid receptors on adipocytes and immune cells, niacin supplementation may prove to be an effective way of alleviating the disease (Mahmodi and Rezaei, 2019).

The effectiveness of soybeans for post-pandemic immunity

Among the various functional food ingredients, soybeans contain functional compounds that contribute to health benefits. According to research conducted by Lee et al. (2007), due to host resistance to cancer occurrence or viral infection in mice as the object research is primarily involved with Th1 cell-mediated immunity, the augmented Th1 response in response to the administration of DJ or CGJ fermented soybean products suggests a certain effector function of these fermented soybean products on T helper cell-mediated immune surveillance against infection or oncogenesis. Soybeans contain many biologically functional components, including isoflavones, unsaturated fatty acids, and bioactive peptides, and the fermentation process is known to improve the nutritional and functional properties of soybeans. Thus, the effectiveness of soybeans for the immune system is high because even when it's fermented or processed, it will improve the functional properties, vitamins, and nutrition of soybeans.

Based on the research investigated by Choi et al. (2014), in which they tested the major bioactive compounds such as isoflavones-glycosides in some traditional foods, they found that components of soybeans could affect the immunostimulatory activity at different rates. The immunostimulatory activity of isoflavone aglycones treats immune cells similarly or higher than the immunostimulatory activity with different isoflavone aglycones.

CONCLUSIONS

Soybean is an alternative food for boosting human immune systems. The nutrients in soybeans, such as vitamins, have an abundance amount in them. The vitamins mentioned are A, B1, E, D, and C that can be recognized. There are many vitamins in soybean that are rarely found due to their function to be processed. To enhance the vitamin content of soybean, there are several ways to achieve it; one is the fermentation process. During the fermentation of soybean, vitamin B12, barely found in each soybean, can be enhanced to enough content for human needs. The major bioactive compounds such as isoflavones-glycosides in soybeans could affect the immunostimulatory activity at different rates that treated the immune cells. This stated that soybeans are eligible to be called immune booster food. It is

a recommended choice to consume soybeans daily to prevent viruses or any threats of disease. The act of self-prevention to boost the immune system in the human body can help fight the outbreak such as COVID-19 or similar threats, especially in the post-pandemic situation, so that humans are prepared for health conditions and decrease the mortality rate.

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Effect of wild yam root preparation on physicochemical properties of ready-to-use wild yam flour

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Received: March 29, 2021. Revised: May 6, 2021. Accepted: May 24, 2021

ABSTRACT

Asiatic bitter yam (*Dioscorea hispida* Dennst.) or kloy kao niaw (KKN) is a food plant of people in the tropics, but certain wild varieties are edible after detoxified. Detoxified of KKN are delicious food ingredients. Thus, the effects of the preparation method on the physicochemical quality of ready to used KKN flour were investigated. The KKN tubers were taken from Mae-Tha, Lampang, Thailand. The KKN was cleaned, peeled, and sliced to 0.1 mm before soaked in salt solution at 0, 10, 20, 30, 40, and 50% for 12 hours. Each treatment was washed every day for eight days or until the sliced Kloy has white color. The ready to used KKN flour was produced with three methods; soaked in 2% salt solution for 10 min, boiling for 10 min, and steaming for 30 min prior drying in tray dried at 60 °C for four h then ground through 80 mesh screen and packed in an aluminum foil bag. The ready-to-use KKN flour was analyzed on % production yield, moisture content, color value, gelatinization temperature, starch granule by SEM, and pasting behaviors. Research revealed that the treatment on soaked slice KKN in 20% salt prior steam for 30 min had the highest production yield (26.8%), 7.70% moisture content. The color L* a* b* value were 92.98, 0.15, 7.42 respectively. The selected ready to use KKN flour had low gelatinization temperature, swollen starch granules, and lowest peak viscosity, and easily soluble in water than the other two treatments.

Keywords: table salt solution, detoxified, gelatinization temperature, viscosity behaviors

INTRODUCTION

Asiatic bitter Thai yam (*Dioscorea hispida* Dennst. var. *hispida*), is locally known as kloy kao niaw (KKN) and kloy kao choa (KKC). The tuber is poisonous because of high content of the alkaloid dioscorine and diosgenin. The toxic alkaloid, dioscorine, and diosgenin are poisonous to the central nervous system hence the danger to consumers (Kang et al., 2012; Hudzari et al., 2011). Traditional Thai dishes from KKN and KKC were taken time to prepare for food. The traditional processing is done by slicing the tuber very thinly and soaking it in brine for at least three days then, in flowing water or river for an additional four days until the yam slide had white color. After this, the yam chips are boiled or steamed before eaten, similar to other root crops. The drying process involving chopping, washing, and drying of the tuber starch also reduces some toxins (Kresnadipayana and Waty, 2019). In the case of highly toxic *Aconitum carmichaelii* Debeaux traditionally eaten by the locals of Qinling Mountains (China), detoxification of the root vegetable requires drying and long hours of boiling up to 8-10. Recently, the DNA barcoding strategy

could be used to accurately identify the plant in the wild, which could be labeled with appropriate warnings together with preparation guides to avoid incidences of plant food poisoning (Buenavista et al., 2021). Previously studied by Jayakody et al. (2009) on the ratios of KKN to salt at 1:10 2:10 3:10 4:10 and 5:10 at room temperature for three days. The control was unsalted KKN. Each treatment was rinsed six times per day at 4 hours intervals and dried in an oven at 60 °C for 24 hrs. The dried KKN was diluted with distilled water to 10 100 200 300 and 400 g/L and mixed in water the keep ten tilapia at room temperature for 96 hours. They found that the KKN provided the lowest toxicity to tilapia with 96 h-LD50=204.54 and 214.29 g/L, respectively. For 400 g/L concentration, detoxified KKN with 4:10 and 5:10 ratios showed an accumulated mortality rate of 6.66±3.33 and 3.33±3.33%, respectively. They also reported the approximate composition of the KKN and KKC tuber per 100 g edible portion is: water 78 g, protein 1.81 g, fat 1.6 g, carbohydrates 18 g, fiber 0.9 g, and ash 0.7 g. On a dry weight basis, the tuber also contains 0.2-0.7% diosgenin and 0.044% dioscorine; these poisons can cause paralysis of the central nervous system. Commercial starch extracted from the tubers contains 88.34%

starch, 5.28% protein, 5.33% fiber, 0.23% fat, 0.66% ash, and various phenolic compounds (Kresnadipayana and Waty, 2019; Irfa et al., 2019). Furthermore, Anuntagool et al. (2006) proposed that the Starch from Thai yam *Dioscorea hispida* Dennst; locally known as kloy kao niaw (KKN), was hydrothermally modified (HM) at moisture contents (MC) between 13 and 30 g water/100 g starch (wb) at 90 °C for ten h. All treatments caused a decrease in swelling power and amylose leaching and an increase in gelatinization temperature of the starch, indicating a strengthened network within the starch granule. They also report that KKN starch granule's crystalline structure changed from B to C-type when modified at 13 g water/100 g starch (wb) and stayed unchanged when modified at 18 to 30 g water/100 g starch (wb). Thus, the objective of this study was to investigate the effects of the preparation method on the physicochemical quality of ready to used KKN flour in various food products.

MATERIALS AND METHODS

Kloy kao niaw (KKN) (*Dioscorea hispida* Dennst.) tubers were obtained from Mae-Tha, Lampang Thailand. The KKN roots were stored at room temperature for two days before produce ready to used KKN flour. Table salt was purchased from a local market in Muang Lampang province, Thailand. The chemicals used were all analytical grade from Merck, Germany. All equipment used from the Food Innovation Center, Agricultural Research Institute and Department of Argo Industry, Faculty of Science and Agricultural, Rajamagala University of Technology.

De-toxic of KKN tuber: The toxic alkaloid, dioscorine, and diosgenin were removed following the method from (Pamutha and Kansarn, 2014) with a slight modification. The raw KKN tubers were cleaned under running tap water to removed dirt and soil residues. The cleaned KKN tuber was peeled and sliced to 0.1 mm. The slices KKN were washing and soaked in salt solution at 0, 10, 20, 30, 40, and 50% for 24 hours at room temperature. The treatment was washed three times, and change the salted solution every day until the KKN sliced became white color, and the soaked water had total soluble solid less than 0.05 °Brix and pH within 5-7. Each of the treatments was repeated with the same salted solution for eight days. The drained de-toxic KKN slice was dried at 50 °C for 4 hours before analysis on moisture content (%), production yield (%), and color value.

KKN flour preparation: The ready-to-use KKN flour was produced following Anuntagool et al. (2006), modified using the de-toxic KKN sliced. The KKN flour was prepared by 1) soaked of the de-toxic KKN sliced in 2% salt solution for 10 min, 2) boiling de-toxic KKN sliced for 10 min, and 3) steaming of de-toxic KKN sliced for 30 min. The three preheated de-toxic KKN chips were drying in a tray drier at 60 °C for four h. The dried KKN chips were ground through 80 mesh screens in a Cycotex sample mill (Foss, German) and packed in air-tight aluminum foil bags for future physicochemical analysis as follows.

KKN flour yield: The production yield of the KKN flour was calculated by dividing the original mass of the rough KKN tuber (g) before processing to KKN flour weight (g) and multiple by 100. Moisture content (%) was measure in the Direct Heating Method (AOAC, 2002). Color values of KKN flour samples were measured in triplicate by reflectance using a colorimeter (Color Quest, Hunter Lab). Color value was measured following three parameters: lightness /brightness or whiteness, (L^*) in which black is no reflection and white is perfect diffuse reflection; greenness to redness (a^*), in which negative values indicate green and positive values indicated red and blueness-yellowness (b^*), in which negative values indicate blue and positive values indicate yellow.

Granule morphology: Each KKN flour sample was sprinkled on the masking tape. The images of the KKF starch granules mounted on a stub and gold-coated were recorded using a Scanning electron microscope (JEOL model JSM-6400, Tokyo, Japan) operating at 20 kV with 5,000x magnification following Egerton (2000) with modification.

Pasting properties of KKN flour: The pasting properties of the 3 KKN flours was measure by the Rapid Visco Analyzer Series 4 (RVA-4) Newport Scientific Pty. Ltd., Australia) according to the procedure described by Approved Method 26-21A (AACC, 1999). Briefly, 3 g (12% moisture content) of each sample was mixed with 25 mL of distilled water. Each sample then underwent controlled heating and cooling cycles with constant shear, where it was held at 50 °C for 1 min, heated from 50 to 95 °C at a rate of 6 °C/min, then held at 50 °C for 5 min. The rotation speed was maintained at 75 rpm. The pasting properties, including peak viscosity (PV), hold strength (HS), breakdown (BD = PV-HS), final viscosity (FV), and setback (SB = FV-PV) of all samples were tested in triplicates.

Statistical Analysis: Completely randomized design (CRD) was used to evaluate the means of the physicochemical analysis of kloy samples. The data obtained were subjected to analysis of variance (ANOVA), followed by Duncan's multiple range test procedures for differences between treatments using Minitab for Windows Release 16.1.

RESULTS AND DISCUSSION

Physicochemical properties of the de-toxic KKN chips

Physicochemical properties of the de-toxic KKN chips that were soaked in salt solution at 0, 10, 20, 30, 40, and 50% for 24 h for eight days are shown in Table 1. Results revealed that the production yield of the de-toxic KKN chips treatment with 20% salted solution had the highest production yield (25 %), which was 10.33% moisture content. In addition, the color value of the de-toxic KKN chip

had the whiteness (L^*) value were in the range of 91.8 to 93.4, which was increased by the higher salt concentration. While redness (a^*) value were decreased up on higher % salt solution, which was 0.36 reduced to -0.51. Furthermore, the b^* value also decreased from 7.70 to 6.04. This due to the alkaloid dioscorine and diosgenin was dissolved in the salted solution that became a yellowish color during washing every day within eight days. Thus, the detoxing of KKN tuber sliced with brine (40% salt solution) and follow by watching every day was increased on L^* value of the detoxined KKN chips, which was higher (93.04) than the other treatments. However, for the confidence on the KKN flour poisonous, the author will continue to study the toxin contaminated in the KKN flour by a study on DNA barcoding technique that which was proved to be useful in the accurate and correct identification of *Dioscorea hispida* Dennst as investigated by (Buenavista et al. (2021).

Table1. Physicochemical properties of de-toxic KKN chips soaked in a salt solution for eight days

De-toxic KKN properties	De-toxic KKN					
	0% salt	10% salt	20% salt	30% salt	40% salt	50% salt
Yield (%)	19±0.10 ^b	14±0.40 ^d	25±0.24 ^a	18±0.24 ^b	16±0.60 ^c	13±0.60 ^e
Moisture (%)	11.6±0.1 ^a	10.63±0.1 ^b	10.33±0.3 ^b	9.81±0.1 ^{bc}	10.63±0.2 ^c	10.89±0.5 ^b
Color value L^*	92.60±0.5 ^a	91.98±0.17 ^c	92.98±0.18 ^a	92.92±0.04 ^a	93.04±0.21 ^a	92.46±0.2 ^b
a^*	0.36±0.01 ^a	0.28±0.03 ^a	0.15±0.01 ^c	0.22±0.00 ^b	0.13±0.02 ^c	-0.51±0.02 ^e
b^*	7.70±0.09 ^a	7.40±0.14 ^b	6.41±0.14 ^c	6.43±0.09 ^c	6.50±0.25 ^c	6.04±0.08 ^d

^{a, b} Values are mean ± standard deviation. Different superscripts in columns differ significantly ($P < 0.05$)

^{ns} Values are mean ± standard deviation in columns not significantly ($P > 0.05$)

Physicochemical properties of KKN flour

Analysis results on flour yield, moisture content, color value, granule morphology, pasting behaviors are presented in Table 2 and Figure 1 – 2. Results KKN flour yield revealed that the 2% salt-soaked had the highest percent yield, moisture content, a^* value, pasting time, and gelatinization temperature, 28.15%, 9.88 %, 0.23 a^* , 6.45 b^* , 5.25 min, and 89.90 °C, respectively. In addition, the 30 minutes steamed KKN flour was the lowest value of moisture content, a^* and b^* value, pasting time, and gelatinization temperature as 26.86%, 7.70%, -0.54 a^* , 6.14 b^* , 4.00 min, and 64.53 °C, respectively.

The pasting time and temperatures of raw KKN flour prepared by soaked in 2% salt for 10 min observed by RVA were 5.25 min and 89.90 °C. These results are higher than the previous study, about 78.4-79.6 °C (Naksriarporn et al., 2005). These results indicate that a hard granular structure of raw KKN flour compared to the boil and steamed KKN chip was 66.43 and 64.53 °C. Thus, the 30 minutes steamed KKN flour had good properties for ready to used KKN flour in various food products such as bakery, snack, and breakfast cereal products.

Table2. Physicochemical properties of ready to used KKN flour with three preparation methods.

Physicochemical properties	Preparation methods		
	10 minutes 2% salt-soaked	10 minutes boiled	30 minutes steamed
KKN flour yield (%)	28.15 ±0.10 ^a	26.74±0.12 ^b	26.86±0.62 ^b
Moisture content (%)	9.88±0.01 ^a	8.32±0.02 ^b	7.70±0.02 ^c
Color value L*	92.92±0.53 ^{ns}	92.86±0.01	92.27±1.15
a*	0.23±0.00 ^a	0.15±0.04 ^a	-0.54±0.06 ^b
b*	6.45±0.01 ^{ns}	6.78±0.03	6.14±0.02
Pasting time (min)	5.25±0.26 ^a	4.50±0.24 ^b	4.00±0.12 ^b
Gelatinization temp (°C)	89.90±1.51 ^a	66.43±0.49 ^b	64.53±0.80 ^c

^{a, b} Values are mean ± standard deviation. Different superscripts in rows differ significantly ($P < 0.05$)

^{ns} Values are mean ± standard deviation in columns not significantly ($P > 0.05$)

Granule morphology of KKN flour

Figure 2 shows the images of ready to used KKN flour granule viewed at 5,000 x g magnification viewed under SEM. It was evident that the 2% salt-soaked had granules are mainly polyhedral in shapes with a smooth surface of 1-6 μm . Their shapes and sizes are averages of 3 to 8 μm , and these results are in agreement with a previous study by Santhanee et al. (2014). The smooth surface of the granules of these starches indicated that the treatment with the 2% table salt solution used in this study did not damage the starch granules. While the 10 minutes boiled KKN flour was partially gelatinized starch

granule, which was swelled starch granule. In addition, the 30 minutes steamed KKN flour was partially gelatinized without starch granules shape. Thus, the shared morphological properties of the 30 minutes steamed KKN flour indicated that the steamed process was affected the overall structure of KKN starches. Furthermore, it has been reported that the appearance of starch granule may affect its physicochemical properties, such as gelatinization, pasting, enzyme susceptibility, crystallinity, and solubility of the flour and starch Lindeboom et al. (2004).

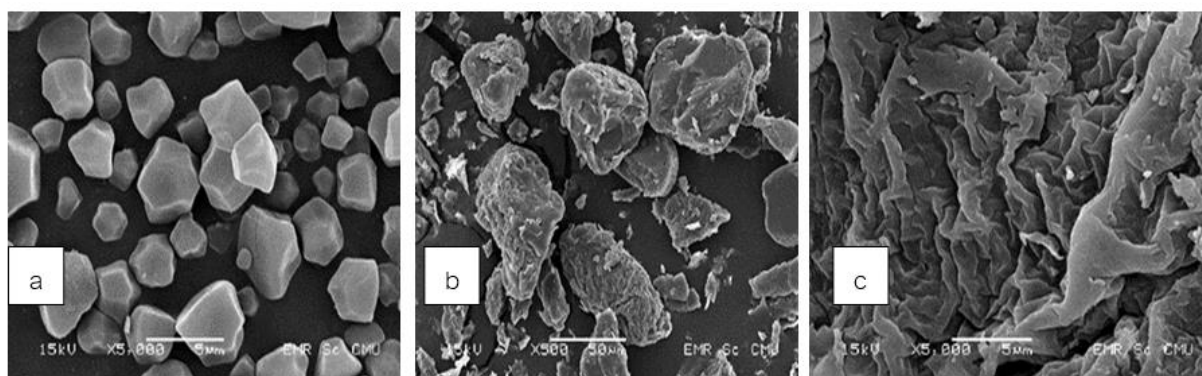


Figure 1. KKN starch granules observed under a scanning electron microscope (a) 2% salt-soaked, (b) 10 min boiled, and (c) 10 min steamed of raw de-toxin KKN tuber.

Pasting behaviors

The pasting behavior on peak viscosity, hold strength, final viscosity, setback, and pasting temperature is shown in Figure 2. The pasting properties of ready to used KKN flour prepared by 2% salt-soaked, 10 min boiled and 10 min steamed of raw de-toxin KNN slice tuber. The pasting curves reflect the behavior of KKN flour during cooking (Mishra and Rai, 2006). The KKN flour prepared from 10 min steamed of raw de-toxin KNN slice tuber GBR was the first to swell and gelatinize due to water uptake at 66.00 °C, followed by 10 min boiled at 82.30 °C and finally, 2% salt-soaked at 89.87 °C

with a statistically significant difference between the three treatments. The 10 min steamed KKN flour had the lowest peak viscosity (87.30 RVU), while the 2% salt-soaked KKN flour showed the highest (392.04 RVU) with 10 min boiled of KKN flour somewhere in the middle (376.09 RVU), which were all significantly different. Our study, therefore, suggests that partially gelatinized KKN flour decreases peak viscosity. Additionally, setback value is defined as the degree of re-association between the starch molecules, is the secondary increase in viscosity during cooling, which eventually determines retrogradation of flour and starch.

The 2% salt-soaked KKN flour had the highest setback (50.10 cp) and was followed by the 10 min boiled of KKN flour (40.0 cp). While the 10 min steamed, KKN flour was the lowest (4.0 cp). This phenomenon is thought to be related to the gelatinization temperature and dispersion of the amylose chain in starch polymers. Mishra and Rai (2006) also suggested that setbacks may largely be determined by the degree of amylose polymerization. From this study, partially gelatinized affected setback, though this cannot be related to the amylose content since the amylose content of KKN flour was significantly different in setback value. Similarly, the breakdown of the different KKN flour preparation revealed that they were significantly different with

the same pattern as viscosity. 2% salt-soaked and 10 min boiled KKN flour had a higher breakdown (92.0 and 85.0 cp) than the 10 min steamed KKN flour (26.30 cp). The degree of gelatinization of KKN flour can be determined using an RVA, based on a calibration curve obtained by proportional mixing of raw and fully gelatinized starch. The best indicator was found to be peak viscosity which decreased with an increase in the degree of gelatinization. A 10 min steamed of raw KKN chips was sufficient to produce completely gelatinized KKN flour. This finding could bring about the application of KKN flour as a ready-to-cook flour and thickening agent in retort foods or foods that require heat-stable viscosity.

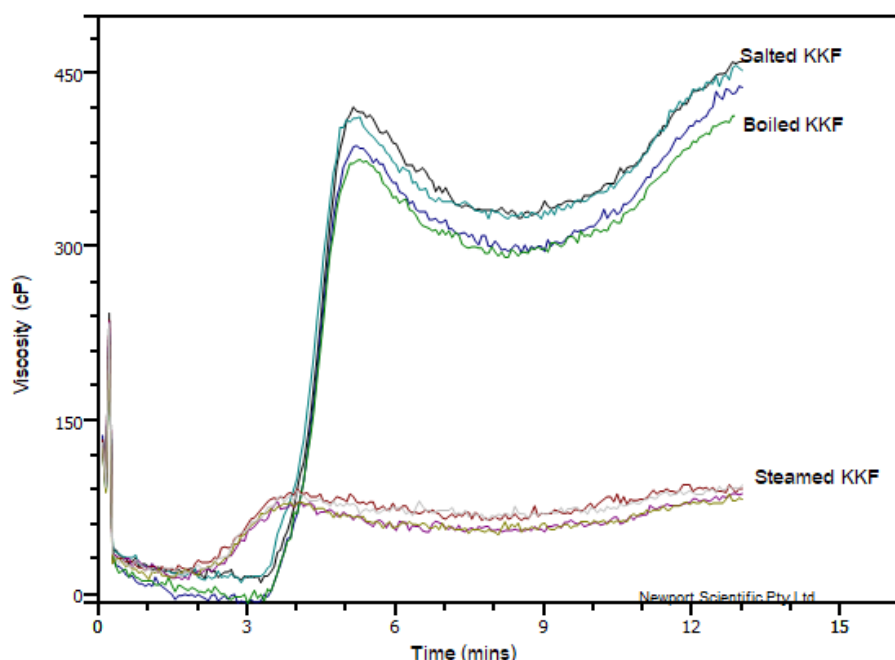


Figure 2. Pasting behaviors of the three treatments of KKN flour production as observed by Rapid Visco Analyzer

CONCLUSIONS

This study showed that the physiochemical of the ready to used KKN flour could be distinguished based on their processing. The quality of the KKN flour of the three treatments even varies the preheated types noticeably. As reported in previous studies by other authors, the shape of starch granules in uncooked KKN flour (*D. hispida*) is dominantly polyhedral and becomes gelatinized with cooking types. The pasting behaviors of the steamed KKN flour could be recommended as a source of ready-to-cooked food industrial raw materials in various products.

ACKNOWLEDGMENTS

The authors are grateful to the Director of Agricultural Research Institute, Rajamangala University of Technology Lanna, for supported research fund within the Plant Genetic Conservation Project under the Royal Initiation of Her Royal Highness Princess Maha Chakri Sirindhorn (RSPG)

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Phytochemical contents and antioxidant activities of Thai herbal tea from leaves of *Morus alba* and *Citrus hystrix*

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Received: March 15, 2021. Revised: April 22, 2021, May 17, 2021. Accepted: May 18, 2021

ABSTRACT

A herbal tea composed of *Morus alba* L. (leaves); *Citrus hystrix* DC. (leaves); 1: 1 (w:w) was extracted using aqueous media. Phytochemicals screening determined total phenolic content (TPC), total flavonoid content (TFC), and the identification of flavonoids and phenolic acids of the extract using High-Performance Liquid Chromatography (HPLC). Antioxidant activities, including 2,2-diphenyl-1-picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) were also assayed. The results showed that the TPC and TFC of the extract were 8.18 ± 0.26 mg GE/g Ext and 6.79 ± 0.75 mg RE/g Ext, respectively. The HPLC analyses of flavonoid and phenolic compounds indicated the presence of catechin, myricetin, quercetin, kaempferol, gallic acid, syringic acid, and coumaric acid as bioactive compounds. Antioxidant activities for DPPH and FRAP were 3.51 ± 0.03 mg TE/g Ext and 15.64 ± 0.36 mM Fe²⁺ E/g Ext, respectively. This finding indicates that Thai herbal tea derived from leaves of *M. alba* and *C. hystrix* has an inhibitory free radical effect and has the potential to promote health following herbal tea consumption.

Keywords: herbal tea, *Morus alba*, *Citrus hystrix*, phytochemistry, antioxidant

INTRODUCTION

Free radicals are one of the causes of many illnesses in humans, especially chronic diseases such as cancer, heart disease, cerebrovascular disease, hypertension, kidney disease, cataract, and gout through multiple mechanisms (Chan and Lim, 2006). Generally, reactive oxygen species (ROS) are a class of compounds that are formed from oxygen metabolism. These highly reactive molecules, such as the hydroxyl radical ([•]OH), peroxide (ROO[•]), and superoxide radicals (O₂^{•-}), can cause severe damage to cells and tissues during various diseases which are linked to heart disease, carcinogenesis, and many other health issues. Synthetic antioxidants such as butylated hydroxyl anisole (BHA), propyl gallate (PG), butylated hydroxyl toluene (BHT), which have been used to prevent oxidation, have been found to cause internal and external bleeding in rats and guinea pigs at high dose (Iqbal et al., 2015). Nowadays, researchers are interested in using natural antioxidants such as bioactive flavonoids, which are of great importance due to their indigenous origin and strong efficacy to trap and scavenge free radicals (Borneo et al., 2009). In recent years, numerous studies have reported that phytochemicals from

plants have antioxidant potential, which are risk factors for many diseases (Duthie and Crozier, 2000).

Herbal teas (more accurately known as tisanes) are mixtures of dried leaves, seeds, grasses, nuts, bark, fruits, flowers, or other plant parts that give them their flavor and provide therapeutic benefits to the body form of herbs (Ravikumar, 2014). The herbal tea in this study was a combination of dried leaves consisting of *Morus alba* and *Citrus hystrix*. This herbal tea developed by Kingyok Registered Ordinary Partnership, Borabue District, Maha Sarakham Province, Thailand (Kingyok ROP) as an alternative to conventional health care, with effects such as lowering blood sugar levels lowering blood lipids, lowering blood pressure, helping relaxation and detoxifying the body

M. alba (Family: Moraceae) known as mulberry, is a deciduous tree widely cultivated in subtropical, tropical, and temperate environments (Mahmoud et al., 2017). It has long been used in traditional medicines in many countries, including Thailand (Soonthornsit et al., 2017). The leaves have been used in Thai traditional medicine as an antipyretic, antitussive, sedative, antidiabetic, and to improve eyesight. Phytochemical composition in mulberry leaves consists of terpenoids, alkaloids, chalcones (including morachalcones B, C),

flavonoids (including astragalin, cyclomulberrin, isoquercitrin, kaempferol, morusin, quercetin, rutin, roseoside, and scopolin), anthocyanins, phenolic acids (including m-coumaric acid, syringic acid, and vanillic acid), stilbenoids (including mulberrosides A, B, F) and coumarins (Chan et al., 2016). Several recent studies have shown that mulberry leaves have antioxidant properties (Thabti et al., 2012), α -glucosidase inhibitory activity (Hwang et al., 2016), antidiabetic (Hansawasdi and Kawabata, 2006), antianxiety (Yadav et al., 2008), anti-inflammatory (Choi and Hwang, 2005), anti-atherosclerotic, anti-obesity, hypolipidemic effects, cognitive-enhancing effects and skin-whitening properties (Chan et al., 2016).

C. hystrix (Family: Rutaceae) is a native species in Asia, especially in tropical regions, and known as Kaffir lime. It is evergreen, aromatic, and distinctive with double-shaped compound leaves (Ratseewo et al., 2016). The leaf of Kaffir lime is a common spice used as a condiment in various Thai and Malaysian recipes (Butryee et al., 2009). In Thailand, it is used in traditional medicine by using leaves as a medicine antitussive, expectorant and carminative. The important biological constituents found in kaffir lime leaves are volatile compounds such as citronellal, linalool, β -cubebene, β -pinene, myrcene, limonene, γ -terpinene, p -cymene, terpinolene, copaene, caryophyllene, citronellyl acetate, citronellol, geranyl acetate and δ -cadinene (Lawrence et al., 1971). In addition, it contains coumarins, alkaloids, carbohydrates, flavonoids, glycosides, phenols, steroids, and tannins (Ali et al., 2015). Pharmacological effects of Kaffir lime leaves have been shown to include antioxidant (Ratseewo et al., 2016), hepatoprotective effect (Abirami et al., 2015), and anti-cancer activity (Tunjung et al., 2015).

This herbal tea was further developed by Kingyok ROP to select a herbal tea for health care. This study performed phytochemical screening (using TPC, TFC, and HPLC) and assayed for antioxidant activity (DPPH and FRAP assay) as a preliminary pharmaceutical investigation.

MATERIALS AND METHODS

Plant material and extraction

The herbal tea was composed of *M. alba* L. (leaves) and *C. hystrix* DC. (leaves); mixed in ratio 1:1 (w:w). Herbal tea was supplied by Kingyok ROP. All the fresh materials were cleaned and dried at 45 °C for 48 h in a hot air oven and then powdered. For extraction (Ext), 1 g of tea in powder form was extracted by immersing in 75 ml water (1:75 w/v) at 80 °C for 15 min. Extracts were filtered through a 0.45 μ m membrane filter. The extracts were separated

into two parts. Part 1 was stored at 4 °C for analysis using TPC, TFC, DPPH, and FRAP assay, which were compared with a single tea (*M. alba* leaf tea and *C. hystrix* leaf tea). Part 2 was stored at -18 °C for analysis by HPLC (Nammatra et al., 2021).

Total phenolic content (TPC) determination

Total phenolic content was determined according to a modified procedure (Singleton et al., 1999). The extract from herbal tea (100 μ L) was oxidized with 500 μ L of 0.2 N Folin-Ciocalteu's reagent and neutralized by adding 400 μ L of 7.5% Na_2CO_3 . The absorbance was measured at 765 nm by UV-Vis Spectrophotometer after mixing and incubated at room temperature for 30 min. The results were expressed as gallic acid equivalent (mg GAE/g Ext).

Total flavonoid content (TFC) determination

Flavonoid content was estimated using the aluminum chloride colorimetric method (Sharma and Agarwal, 2015). The extract from herbal tea (500 μ L), 2,000 μ L distilled water, and 150 μ L 5% NaNO_2 solution were added. After 6 min, 150 μ L 10% AlCl_3 solution was added and kept for another 6 min. To this reaction mixture, 2,000 μ L 4% NaOH solution and 200 μ L water were added to make up the final volume of 5,000 μ L. The reaction mixture was mixed well and allowed to stand for 15 min after which absorbance was recorded at 510 nm. The total flavonoid content (TFC) was calculated from a standard rutin equivalent (mg RE/g Ext).

High-Performance Liquid Chromatography (HPLC) analysis

Analysis to compare the amount of some flavonoid and phenolic acid compounds from herbal tea that was beneficial to the health used the High-Performance Liquid Chromatography (HPLC) method according to Jorjong et al. (2015). HPLC–DAD system (Shimadzu, Japan), comprising of Shimadzu LC-20AC pumps, a SPD-M20A diode array detector, and an Apollo C-18 column (Alltech Associates, Deerfield, IL, USA) (4.6 mm x 250 mm, 5 μ m) protected with guard column Inertsil ODS–3 (4.0 mm x 10 mm, 5 μ m; GL Science Inc., Tokyo, Japan) were used for the analysis of flavonoids and phenolic acids. The mobile phase for flavonoids used acetonitrile/deionized water (2/97.8, v/v) containing 0.2% phosphoric acid (solvent A) and acetonitrile/deionized water (97.8/2, v/v) containing 0.2% phosphoric acid (solvent B) at a flow rate of 0.6 ml/min and 40°C column temperature. The UV–Vis spectra were detected at 254 nm. (Butkhup and Samappito, 2008). The mobile phase for phenolic acids consisted of acetonitrile (solvent A) and

phosphoric acid in deionized water pH 2.58 (solvent B) at a flow rate of 0.8 ml/min and 40 °C column temperature. The flavonoid compounds detected were catechin, myricetin, quercetin, and kaempferol, and the phenolic compounds were gallic acid, syringic acid, and coumaric acid, and all were analyzed by comparing the retention time and spectrum as well as standard addition.

Antioxidant activity by DPPH assay

2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging capacity of extract from herbal tea was estimated by the reduction of the reaction color of DPPH solution and sample extracts by the method according to Thaipong et al. (2006). DPPH was dissolved in ethanol at 0.039 mg/mL. The extract was diluted with distilled water to yield sample solutions at various concentrations. 100 µL of the sample solution was added to 900 µL DPPH (0.1 mM) to give the working solution. After a 30 min reaction kept in the dark at ambient temperature, the absorbance of the solution was measured at 517 nm. Trolox (6-hydroxy-2,5,7,8-tetramethylchlorman-2-carboxylic acid) was used as a positive control for comparison, and solvent mixed with 0.1 mM DPPH solution was taken as a negative control. The percent scavenging was calculated by the following formula:

$$\text{DPPH scavenging activity (\%)} = [(A_0 - A_s) / A_0] \times 100$$

A_0 of control was the absorbance of the solvent mixed with DPPH solution, and A_s was the absorbance of the extract solution. DPPH radical scavenging was indicated as mg Trolox equivalent (mg TE)/g extraction.

Ferric reducing antioxidant power (FRAP) assay

The FRAP assay was conducted according to a previously described method by Benzie and Strain (1996). The working solution was prepared by mixing 25 mL of acetate buffer pH 3.6 (3.1 g of $\text{CH}_3\text{COONa} \cdot 3\text{H}_2\text{O}$ and 16 mL of CH_3COOH) to a concentration of 300 mM, 2.5 mL TPTZ solution (10 mM TPTZ in 40 mM HCl), and 2.5 mL of 20 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ solution and equilibrated at 37 °C before use. Samples had a concentration of 1500 µg/mL (200 µL) and could react with 2.5 mL of the working solution for 30 min in the dark at 37 °C. Absorbance was measured at 593 nm ($n=3$) using a UV-Vis spectrophotometer. Ferrous sulfate (FeSO_4) was used as a standard to establish a standard curve. The FRAP antioxidant activity was expressed as mM of Fe^{2+} equivalents per g of sample (mM Fe^{2+} /g Ext).

Statistical analysis

All assays were expressed as mean \pm standard deviation (SD) from five separate experiments ($n=5$). Statistical analysis was carried out using one-way analysis of variance (ANOVA) followed by Duncan's multiple range tests. Differences at $P < 0.05$ were considered to be significant.

RESULTS AND DISCUSSION

Total phenolic and total flavonoid contents

The total phenolic contents (TPC) were estimated using gallic acid, and total flavonoid contents (TFC) were estimated using rutin as standard. The results showed that the TPC and TFC of extract from HT were 5.89 ± 0.07 mg GAE/g Ext and 8.38 ± 0.33 mg RE/g Ext, respectively (Table 1), indicating a moderate total phenolic and total flavonoid content in herbal tea extract. These were significantly higher contents than *M. alba* leaf tea (TPC = 5.48 ± 0.18 mg GAE/g Ext and TFC = 7.26 ± 0.45 mg RE/g Ext), but were significantly lower than *C. hystrix* leaf tea (TPC = 20.21 ± 0.06 mg GAE/g Ext and TFC = 12.03 ± 0.24 mg RE/g Ext). Furthermore, other research has reported important total phenolic and flavonoid content in aqueous extracts derived from herbal tea from the leaves of *M. alba* and *C. hystrix*. Thabti et al. (2012) found that the aqueous extract of *M. alba* leaves showed TPC (759 ± 74 mg GAE/100 g DW) and TFC (717 ± 45 mg RE/100 g DW), and these levels were greater than in methanolic extract (TPC = 560.00 ± 97.23 mg GAE/100 g DW and TFC = 283.13 ± 4.10 mg RE/100 g DW). Radojković et al. (2012) reported that the ethanol extract of *M. alba* leaves had TPC at 66.766 ± 0.749 mg CAE/g and TFC at 33.303 ± 0.059 mg RE/g. There is also a report that methanol and ethanol extracts of *C. hystrix* leaves had TPC levels of 1.40 ± 0.32 mg GAE/g and 1.37 ± 0.32 mg GAE/g, and TFC were 2.58 ± 0.71 mg RE/g and 2.30 ± 0.56 mg RE/g, respectively (Ali et al., 2015). In 2016, Ratsewo et al. (2016) reported that the aqueous extracts obtained from steamed *C. hystrix* leaves had TPC at 22.18 ± 0.06 mg GAE/g and TFC was 11.84 ± 0.02 mg RE/g.

Phenolic compounds from plants, such as flavonoids, have antioxidant potential, which is a risk factor for disease (Duthie and Crozier, 2000). Furthermore, phenolic compounds are considered secondary metabolites, and these phytochemical compounds derived from phenylalanine and tyrosine occur ubiquitously in plants and are diversified and are thought to have positive effects on human health (Saeed et al., 2012). The aqueous extraction is a method used to brew tea and not harmful to humans. Nevertheless, aqueous extraction showed a low level

of total phenolics due to most flavonoids being non-polar, making them less readily extractable with water, which is a strong polar solvent (Mojulat and Surugau, 2018). Furthermore, the aqueous extraction may result in lower total phenol levels than other

methods or possess phenolic compounds that contain a smaller number of active groups than the other solvents (Do et al., 2014).

Table 1. Total phenolic contents (TPC) and total flavonoid contents (TFC) of extract from herbal tea and single tea.

Samples	TPC (mg GE/g Ext)	TFC (mg RE/g Ext)
HT	5.89 ± 0.07 ^b	8.38 ± 0.33 ^b
<i>M. alba</i> leaf tea	5.48 ± 0.18 ^{bc}	7.26 ± 0.45 ^c
<i>C. hystrix</i> leaf tea	20.21 ± 0.06 ^a	12.03 ± 0.24 ^a

HT= Herbal tea was composed of *M. alba* L. (leaves) and *C. hystrix* DC. (leaves); mixed in ratio 1:1 (w:w).

TPC was measured with gallic acid equivalents (mg GE/g Ext). TFC was measured with rutin equivalent (mg RE/g Ext).

Values in the columns with different superscript letters are significantly different ($P < 0.05$).

Flavonoids and Phenolic acids

A partial analysis of the flavonoids and phenolic acid compositions of the healthful herbal tea was performed using reversed-phase HPLC. Generally, the same flavonoid and phenolic acid compounds were present in all plant species, but there were differences in relative levels (Jorjong et al., 2015). In this study, partial flavonoid content analysis of the aqueous extract from herbal tea were catechin, myricetin, quercetin, and kaempferol. The results showed that the content of catechin, myricetin, quercetin, and kaempferol were 2324.77 ± 34.43 µg/g Ext, 170.36 ± 5.82 µg/g Ext, 6176.80 ± 11.72 µg/g Ext and 382.16 ± 3.54 µg/g Ext, respectively (Table 2). Previously, research has shown that the leaf extract of *M. alba* contained flavonoid constituents consisting of astragalin, atalantoflavone, cyclomulberrin, isoquercitrin, kaempferol, kaempferol 3-O-β-D-rutinoside, morusin, quercetin, quercetin 7-O-β-D-glucopyranoside, rutin, roseoside, skimmion and scopolin (Chan et al., 2016). The leaf extract of *Citrus hystrix* contains many flavonoids, such as myricetin, peonidin, cyanidine, quercetin,

luteolin, hesperetin, apigenin and isorhamnetin (Butryee et al., 2009).

Gallic acid, syringic acid, and coumaric acid comprised the phenolic acid content of the aqueous extracts. The results showed that the content of gallic acid was higher than syringic acid and coumaric acid, which were 1,053.00 ± 76.30 µg/g Ext, 195.54 ± 9.72 µg/g Ext, and 182.81 ± 11.03 µg/g Ext, respectively (Table 3). Past studies have reported that the leaf extracts of *M. alba* have 5-O-caffeoylquinic acid, m-coumaric acid, p-coumaric acid, ferulic acid, gallic acid, hydroxybenzoic acid, protocatechuic acid, protocatechuic aldehyde, syringaldehyde, syringic acid, and vanillic acid (Chan et al., 2016), caffeoylquinic acid and caffeic acid (Thabti et al., 2012). The extract from the peel of *C. hystrix* has gallic acid, caffeic acid, p-coumaric acid, and ferulic acid (Wijaya et al., 2017). It is possible that the extraction of mixtures of more than one plant species together causes changes in the content and type of phytochemical. This study can indicate that bioactive compounds from leaves might be potential natural sources for the development of antioxidant function in dietary food.

Table 2. Flavonoid content (µg/g Ext) of extract from herbal tea.

Samples	Catechin (µg/g Ext)	Myricetin (µg/g Ext)	Quercetin (µg/g Ext)	Kaempferol (µg/g Ext)
HT	2324.77 ± 34.43	170.36 ± 5.82	6176.80 ± 11.72	382.16 ± 3.54

HT= Herbal tea was composed of *M. alba* L. (leaves) and *C. hystrix* DC. (leaves); mixed in ratio 1:1 (w:w).

Table 3. Phenolic acid content ($\mu\text{g/g}$ Ext) of extract from herbal tea.

Samples	Gallic acid ($\mu\text{g/g}$ Ext)	Syringic acid ($\mu\text{g/g}$ Ext)	Coumaric acid ($\mu\text{g/g}$ Ext)
HT	1053.00 \pm 76.30	195.54 \pm 9.72	182.81 \pm 11.03

HT = Herbal tea was composed of *M. alba* L. (leaves) and *C. hystrix* DC. (leaves); mixed in ratio 1:1 (w:w).

Antioxidant activity

In this experiment, the radical scavenging activities of extract from herbal tea were measured using two different assays, namely DPPH and FRAP, as shown in Table 4. The antioxidant activity in aqueous extract of HT for DPPH was 3.51 ± 0.03 mg TE/g Ext, while FRAP radical scavenging activity of HT gave a moderate antioxidant capacity in this study which showed a reduction of ferrous ion (Fe^{2+}) radical of 15.64 ± 0.36 mM Fe^{2+} E/g Ext. These were significantly higher contents than *M. alba* leaf tea (DPPH = 3.13 ± 0.12 mg TE/g Ext and FRAP = 13.57 ± 1.10 mM Fe^{2+} E/g Ext), but were significantly lower than *C. hystrix* leaf tea (DPPH = 9.39 ± 0.18 mg TE/g Ext and FRAP = 21.21 ± 1.65 mM Fe^{2+} E/g Ext). This was consistent with the analysis of the total phenolic (TPC) and flavonoids (TFC), which found that kaffir lime tea had the highest values of both. Due to phenolic and flavonoid compounds from plants were antioxidant properties (Duthie and Crozier, 2000).

This result related well with the study of Radojković et al., (2012), reporting that an ethanol extract of *M. alba* showed high antioxidant activity with IC_{50} value by DPPH assay of 0.0124 mg/mL. In

the same year, Chan et al. (2012) report that the aqueous extract with microwave drying from tea of shredded leaves of *M. alba* had antioxidant activity by both DPPH (1920 ± 23 mg AA/ 100 g) and FRAP (865 ± 25 mg GAE/ 100 g). Thabti et al. (2012) reported that the aqueous leaf extract of *M. alba* contained antioxidant activity with an IC_{50} value by DPPH assay of 5.59 ± 0.14 mg/mL. According to the study of Butryee et al. (2009) the aqueous extract of boiled *C. hystrix* leaves can be antioxidant both DPPH ($39 \pm 18\%$ RSA) and FRAP (49.61 ± 21.14 $\mu\text{mole Fe}^{2+}$ E/g). Laohavechvanich et al. (2010) reported that the aqueous extract of boiled *C. hystrix* leaves had an antioxidant activity with IC_{50} value by DPPH assay of 11.9 mg/mL. Furthermore, Ratsewo et al. (2016) found that the aqueous extract of boiled *C. hystrix* leaves showed high antioxidant activity with the FRAP method at 583 ± 18.17 $\mu\text{mol FeSO}_4/\text{g}$. Consistent with past studies, the antioxidative properties of flavonoids are due to several different mechanisms, such as scavenging of free radicals, chelating of metal ions, such as iron and copper, and inhibition of enzymes responsible for free radical generation (Ali et al., 2015).

Table 4. DPPH and FRAP radical scavenging activities of extract from herbal tea and single tea.

Samples	DPPH (mg TE/g Ext)	FRAP (mM Fe^{2+} E/g Ext)
HT	3.51 ± 0.03^b	15.64 ± 0.36^b
<i>M. alba</i> leaf tea	3.13 ± 0.12^{bc}	13.57 ± 1.10^c
<i>C. hystrix</i> leaf tea	9.39 ± 0.18^a	21.21 ± 1.65^a

HT= Herbal tea was composed of *M. alba* L. (leaves) and *C. hystrix* DC. (leaves); mixed in ratio 1:1 (w:w).

DPPH radical scavenging activity was used Trolox as a positive control for comparison.

FRAP radical scavenging activity was reducing of ferrous ion (Fe^{2+}) radical from herbal tea.

Values in the columns with different superscript letters are significantly different ($P < 0.05$).

CONCLUSIONS

The aqueous extract of herbal tea from mixed in leaves of *M. alba* and *C. hystrix* have antioxidant activity based on TPC, TFC, DPPH, and FRAP assays. Flavonoid compounds identified by HPLC consisted of quercetin, catechin, kaempferol, and myricetin. Phenolics detected were gallic acid, coumaric acid, and syringic acid. It can be concluded that this herbal tea has antioxidant effects with the potential to be consumed as an herbal tea for health care. However, there is a need to clarify the major active substance, in vivo and in clinical research in the next study, and study-related other biological activities and other extraction methods.

ACKNOWLEDGMENTS

The authors would like to thank the operators of Kingyok Registered Ordinary Partnership for material support toward this project. This research was financially supported by Industrial Research and Technology Capacity Development (IRCT) under the Northeastern Science Park Project by Science Park, Maha Sarakham University, Maha Sarakham, Thailand.

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Monitoring system for water level and soil moisture for rice fields with LoRa communication on a wireless sensor network

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Received: April 11, 2021. Revised: April 28, 2021. Accepted: May 6, 2021.

ABSTRACT

The monitoring system for water level and soil moisture of wetland rice is an important issue for farmers to pay attention to if more than one field is being monitored. The problem is that farmers are still monitoring manually and without proper tools. These problems underlie this research to implement a monitoring system with the application of the concept of wireless sensor networks with LoRa communication. The implementation of LoRa communication because it has low power consumption so it is suitable for applications in open spaces and long communication range. The application of LoRa communication at sensor and gateway nodes so that it can send sensing data. The data will be forwarded to the data center and can be monitored via a web application. Because there are several fields being monitored, this study uses the concept of a wireless sensor network so that it reaches far away distances of monitored rice fields. The results of the first system evaluation are that the system can measure the water level and soil moisture well. Second, it can send sensor data from the sensor node to the client. Finally, the communication performance of LoRa is the successful rate found in packet delivery, which is a total average of 92.72% or good category. Packet loss with an average total of 7.28% or good category. RSSI signal (Received Signal Strength Indicator) with a total average of -95.95 dBm. The system has good packet delivery success, small delivery failures, and good delivery signal strength.

Keywords: LoRa, monitoring systems, wireless sensor networks, rice fields

INTRODUCTION

One of the important factors of rice plant growth is the availability of sufficient water while seeding to rice harvesting (Rosada et al., 2019). Providing water and maintaining sufficient water content is essential because, besides being able to meet the water needs of rice plants, it can also prevent the growth of weeds that will kill rice seeds. In addition, one of the factors that affect agricultural land is soil moisture, water availability that can be affected by rain or irrigation, and weather, which can cause water evaporation on rice fields (Munir et al., 2018). Soil moisture in rice fields in each season has the optimal humidity for rice plant growth, namely wet category humidity for the beginning of the season, slightly wet for the middle of the season, and dry for the end of the season (Munir et al., 2018).

Generally, farmers in monitoring and knowing the water conditions on their agricultural land need to check directly and periodically on environmental conditions and crops according to the experience of the farmers themselves over the years, and farmers also do not have measuring tools to monitor water conditions in the lowland rice system right (Bakhri and Sudaryono, 2016). Apart from

environmental factors, the main problem indirectly checking the condition of the plants is the distance between the rice fields that need to be checked and monitored from one another. The spacing of fields that need to be checked and monitored makes it impossible for farmers to check all crop conditions directly. From these problems, we need a tool that can monitor water conditions on agricultural land, especially the water level and soil moisture periodically, so that the time to provide water for lowland rice plants is not too late. The monitoring tool needed is a rice field monitoring system that can monitor and operate automatically, one of which is to use wireless sensor network technology.

Wireless sensor nodes are also known as WSN. The Internet of Things is also a development of the WSN network concept that connects the WSN network to the data center through internet data communication. Energy consumption is essential for several systems with the WSN concept so that it can use battery power for WSN systems that have relatively small energy consumption. To support relatively small energy consumption, a technology that also has a low energy consumption is needed, such as the LoRa (Long Range) technology that includes the LPWAN network, which requires low

power energy consumption and has high efficiency in terms of transmitting radio frequencies. Compared with Z-Wave, Bluetooth, and WiFi, which have high energy consumption. Meanwhile, when compared to Zigbee, LoRa communication has advantages in range (Ali et al., 2019). Some LoRa systems have a communication range of more than 2 km, subject to adjustments in terms of environment and configuration. However, LoRa itself has a maximum payload size limit of 243 bytes, so it is suitable for use for WSN networks that send text messages because of its relatively small size (Mekki et al., 2018). Even so, LoRa requires a gateway device that can connect the WSN network with the server as a data center. The WSN network interface with the server can use the websocket protocol, which has advantages over HTTP because it is enough to do a one-time handshake and send messages immediately. The websocket protocol can send and receive packets at the same time (Zhang and Shen, 2013). The websocket protocol compared to other protocols such as HTTP has advantages, namely the lack of header usage, CPU usage, memory consumption, and response time (Chika and Esther, 2019).

Monitoring of paddy fields with parameters of water level and soil moisture is indicated as needed. In addition, with the distance of rice fields that need to be monitored, it is crucial to create a monitoring system that can monitor agricultural land so that it is expected to help ease farmers in monitoring the condition of their agricultural land. This research will use the LoRa communication module, which has a far enough range with low energy consumption equipped with ultrasonic sensors and soil moisture to facilitate data acquisition with a long coverage and the number of nodes more than one. The system also requires a websocket protocol to connect the WSN network with the server as a data center

MATERIALS AND METHODS

The first research on irrigation systems in terraced rice fields was using a wireless sensor network that implements a data acquisition system from rice fields using a water level sensor, namely, the HCSR04 ultrasonic sensor. The system is only limited to a microcontroller circuit device, the results of which are displayed on an android device that is connected wirelessly to the NRF24L01 for communication between nodes and uses Bluetooth to communicate on the android then also moves the servo to open the sluice latch. The system testing used is the result of monitoring conducted by ultrasonic sensors and displayed on a microcontroller to detect water levels, servo testing, and processing time to

show the length of time it takes to detect altitude to process data transmission (Rosada et al., 2019).

The second research is the development of a monitoring system used to monitor the physical condition of fish pond water, which uses the LoRa communication protocol. The method used to develop a gateway that can connect with one other sensor node by using the LoRa communication module to one gateway node with the LoRa communication module and then sent to the data center using the HTTP communication protocol on the Wireless Sensor Network. The results of the tests conducted by this study were the successful rate testing of struct data packets sent from the sensor node to the gateway node with a distance of 50, 100, 200, and 400 meters (Adin et al., 2019).

The next research is to analyze the parameters of the LoRa communication used to determine the RSSI results and packet loss obtained. This research analyzes the transmission of LoRa communication data so that RSSI and packet loss values can be determined based on the testing distance. The test in this study resulted in an RSSI that increased with the farther the distance traveled by the transmitted data packet, whereas packet loss has excellent results at distances of 10 meters to 500 meters and poor results at distances of more than 500 meters to 2 km. It can be seen that data are lost in the package sent (Yanziah et al., 2020).

Rice farming land

In agricultural land, several important factors affect the growth rate of rice plants. One of the important factors of rice plant growth is the availability of sufficient water while seeding to rice harvesting. Providing water and maintaining sufficient water content is essential because, besides being able to meet the water needs of rice plants, it can also prevent the growth of weeds that will kill rice seeds. In addition, one of the other factors that affect agricultural land is soil moisture, water availability that can be affected by rain or irrigation, and weather that can cause water evaporation on rice fields (Munir et al., 2018). Soil moisture in rice fields in each season has the optimal humidity for rice plant growth, namely wet category humidity for the beginning of the season, slightly wet for the middle of the season, and dry for the end of the season (Arif et al., 2014).

Initially, paddy farming was conducted by a dryland cropping system with a field system. This technique has proven to be unsuitable for crop yields due to the limited availability of water on agricultural land. Therefore, the technique of stagnating water in paddy fields began to develop because, actually, the rice plant is a semi-aquatic plant that requires a lot of

water. Especially during the reproductive period, the reproductive phase itself starts from tillers to filling the rice seeds. Water stagnation on rice fields, apart from fulfilling the water needs of rice plants, can also be used to suppress weed growth (Bintariadi et al., 2010).

Wireless sensor networks

The wireless sensor node or wireless sensor network is an infrastructure consisting of measurement, computing, and data communication parts so that it can be used to monitor certain environments or events (Sohraby et al., 2007). The wireless sensor network is part of the wireless sensor network, known as WSN. The Internet of Things is also developing the WSN network concept that connects several sensor nodes on the WSN network to the data center through internet data communication. A node is equipped with sensor equipment that is used to determine changes in the surrounding environment with a communication module that is used to communicate between sensor nodes and to other nodes (Dargie and Poellabauer, 2010). Wireless sensor network architecture consists of several sensor nodes (source nodes) that are connected to each other and sink nodes that are connected to the Internet for later access by users. There are two forms of wireless sensor networks, namely, those that use multi-hop and single-hop. If using multi-hop, it will use more than 1 hop between the sensor node and the gateway node. Meanwhile, if using single-hop, it only uses 1 hop between the sensor node and the gateway node. Single-hop itself can use just point to point using one sensor node and one gateway node, while multipoint to point has more than one sensor node (Dargie and Poellabauer, 2010). The way wireless sensor networks work is that the sensor data obtained from several sensor nodes are collected into a sink node. The role of sink nodes in wireless sensor networks is to collect sensor data and have Internet accessibility. Internet of Things (IoT) is a network that connects a system consisting of various physical devices so that they can communicate with each other via the Internet (Patel and Patel, 2016). The application of hardware into IoT can turn the hardware device into a smart device that can communicate with other hardware devices. This hardware device can see, hear, think and make its own decisions according to the algorithms applied to the hardware. IoT itself is a form of M2M (machine to machine) communication, which is often called an intelligent system.

LoRa

LoRa is a wireless technology that has long-distance services, low power consumption requirements, and security in transmitting data for IoT applications with low transmission rates (Lavric and Popa, 2017). LoRa technology has different frequencies of use in each country. The frequencies used for the United States, the European region, and the Asian region are 915 MHz, 433/868 MHz and 433 MHz, respectively.

Websocket

Websocket is a communication protocol between client and server, which is developed to support full-duplex function (Zhang and Shen, 2013). The websocket communication protocol can support two-way communication between the client and server, which can be used to send and receive data.

Websocket itself is similar to TCP, except that it cannot be used as an alternative to TCP. This is because of how many websocket and TCP are in two different network layers with websocket using the TCP transport layer. The following is Figure 2.3 describes how the work or communication flow of the websocket protocol.

General description of the system

A general description of the system will describe in general how the arrangement of this system includes the architecture that is in it. The following is the system architecture in Figure 1.

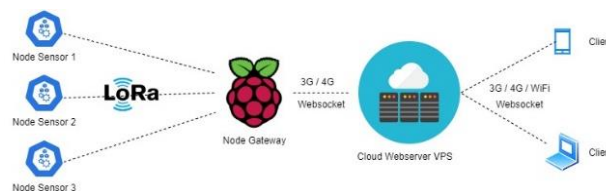


Figure 1. System architecture.

In making hardware design, 3 sensor nodes are needed, with Sensor Node 1 being in the Rice Field 1, Sensor Node 2 being in the Rice Field 2, Sensor Node 3 being in the Rice Field 3. The sensor node consists of a series of microcontrollers in the form of an Arduino Uno equipped with an ultrasonic sensor, which functions to detect water levels in rice fields, a pressure sensor equipped with a tensiometer that detects soil moisture in rice fields, and the LoRa module. In addition, there is also a Raspberry Pi, which is used as a middleware/gateway node that is equipped with a LoRa communication module and a 3G/4G modem with the LoRa communication protocol. All nodes will be connected and communicate with the LoRa communication module

so that they can exchange data in the form of data structs between the sensor and the gateway nodes.

Design of water level sensor nodes

A design is needed to connect the pins between the ultrasonic sensors and the pins on the sensor node microcontroller in the form of an Arduino Uno. Here is picture 2, which describes the sensor pins with the Arduino microcontroller pins.

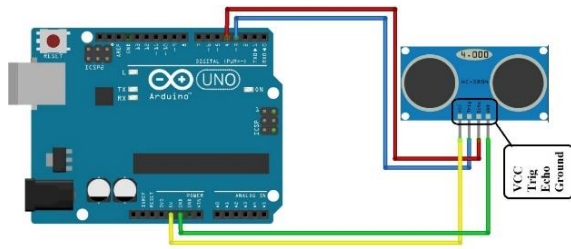


Figure 2. Water level sensor node design.

Soil moisture sensor node design

In addition, a design is also needed to connect the pins between the MPX5700DP pressure sensor and the pins on the sensor node microcontroller in the form of an Arduino Uno. Here is picture 3, which explains the sensor pins with the Arduino microcontroller pins.

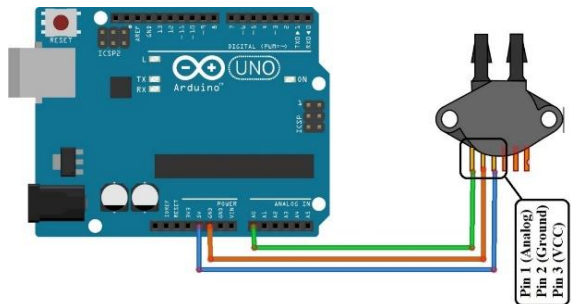


Figure 3. Soil moisture sensor node design.

Sensor node communication design

A design is needed to connect the pins between the LoRa module and the pins on the sensor node microcontroller in the form of an Arduino Uno to be used as a communication module on the sensor node. Here is picture 4, which explains LoRa pins with Arduino microcontroller pins.

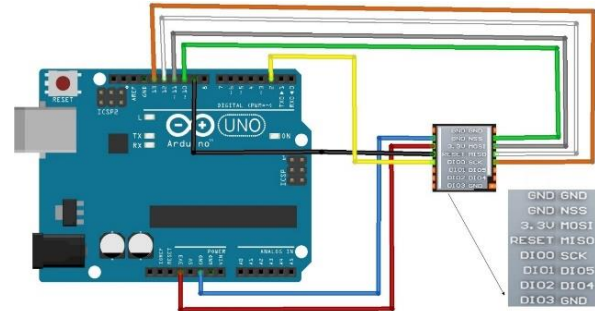


Figure 4. Sensor node communication design.

Communication design node gateway

A design is needed to connect the pins between the LoRa module and the pins on the gateway node microcontroller in the form of the raspberry pi to be used as a communication module at the gateway node. The following is picture 5, which explains LoRa pins with raspberry pi microcontroller pins.

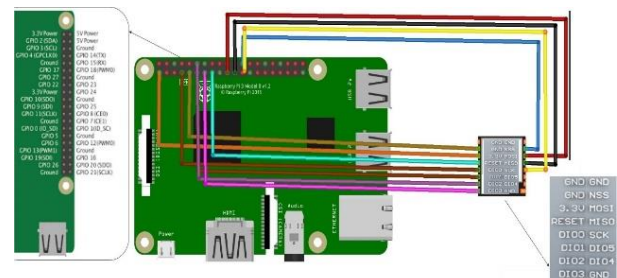


Figure 5. Gateway node communication design.

Server design

The design of a cloud server in this system will be in the form of a web-based application, namely, a web server and client. The cloud server will use Xampp as a server consisting of Apache2 as the webserver plus MySQL as the database. The web server is accompanied by a program with the Python programming language with additional libraries as a server with the websocket communication protocol.

```
New client connected and was given id 1
Data Received :
{'Node': 1}
{'Soil Moisture': '53.57%'}
{'Water Level': '4.00cm'}
Data Sent : Successful
```

Figure 6. Server design.

Sensor node implementation

The sensor nodes in this study consisted of an Arduino Uno as a microcontroller, with two types of sensor modules, namely the HC-SR04 ultrasonic sensor, which is used to detect water levels in paddy fields, and the MPX5700DP air pressure sensor module using a tensiometer used to detect soil moisture based on the recorded air pressure in the tensiometer tube. In addition, there will also be a LoRa SX1278 module which is used for sending sensor data to the gateway node. The implementation of this sensor node is conducted using the Arduino IDE application. The following figure 7 about the sensor node.

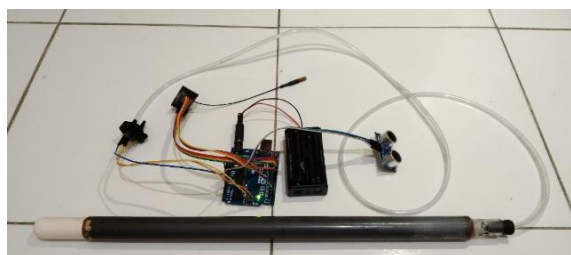


Figure 7. Sensor node implementation.

Gateway node implementation

The gateway node in this study consists of raspberry pi as a microcontroller, with the addition of a modem communication module which is used to get internet accessibility. In addition, there will also be a LoRa SX1278 module which is used to receive sensor data from sensor nodes. The implementation of the gateway node is done using the Python program. The following figure 8 about the sensor node.



Figure 8. Gateway node implementation.

Server implementation

The server implementation is used so that the gateway node can send sensor results to the webserver. The following is a figure 9 implementation of a server using the websocket communication protocol.

```
root@adiwahyucandrakusuma:/opt/lampp/f
New client connected to id 1
New client connected to id 2
Node 1
Soil Moisture 85.59%
Water Level 3.20cm
Data successfully sent to all clients
```

Figure 9. Server implementation.

RESULTS AND DISCUSSION

Water level testing results

Through this test, it was found that the sensor nodes can acquire water level data from the HC-SR04 ultrasonic module. The following is a table of ultrasonic sensor accuracy.

Table 1. Results of water level testing

Testing	Average Error	
Sensor Node 1	Test 1	1, 33%
	Test 2	0, 67%
	Test 3	0, 67%
Sensor Node 2	Test 1	1,33%
	Test 2	0, 67%
	Test 3	0, 67%
Sensor Node 3	Test 1	0, 67%
	Test 2	0, 67%
	Test 3	0%
Average	0, 74%	

The water level data acquisition test for the sensor node is going well, with the testing treatment according to predetermined stages showing that the ultrasonic sensor module can detect the water level value according to the value in reality, which has a fairly high-value accuracy. The water level value obtained is a number in "cm" units. The values recorded in each sensor node can be the same or not the same; this is because the rice fields are sensed differently. Sensor nodes can acquire groundwater level data through the ultrasonic sensor module; the ultrasonic module itself can also provide good data accuracy with an average total error of 0.74% in detecting water level values that match the reality.

Soil moisture testing results

Through this test, the results show that the sensor node can acquire soil moisture data from the MPX5700DP pressure sensor module, which has been connected to a tensiometer. The following are

the results of the accuracy of the MPX5700DP pressure sensor module.

Table 2. Results of soil moisture testing

Testing	Average Error	
Sensor Node 1	Test 1	0%
	Test 2	0%
	Test 3	0%
Sensor Node 2	Test 1	0%
	Test 2	0%
	Test 3	0%
Sensor Node 3	Test 1	0%
	Test 2	0%
	Test 3	0%
Average	0%	

Through testing the soil moisture above, it can be concluded that sensor node 1, sensor node 2, sensor node 3 can acquire data well. The results of each sensor node can be different because the sensor nodes are placed on different rice fields. The water level value obtained is a number in "percent (%)" units that have been calculated and converted from the kPa value obtained by the sensor node. The final results can also provide good data accuracy with an average total error of 0% in detecting soil moisture values that match the reality.

Communication testing results

Through this test, the results show that the gateway node has also succeeded in receiving water level and soil moisture data which have been combined in structured data sent by each of the three sensor nodes via LoRa communication. Here is a picture of 10 terminals on the gateway node.

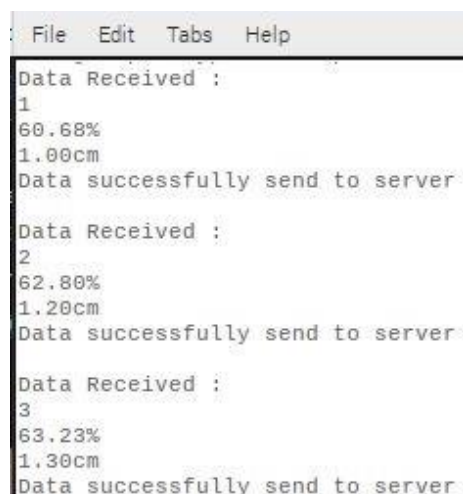


Figure 10. Receiving LoRa data on the gateway.

Testing of each sensor node can send water level and soil moisture data in the form of data structs to the gateway node via LoRa communication and prove that the gateway node can receive water level and soil moisture data in the form of struct data sent by each sensor node via LoRa communication. Three data are sent, the first is the node name data, the second is the soil moisture data, and the last is the water level data.

Websocket data communication testing results

Through this test, the results show that the vps cloud server can receive water level and soil moisture data which have been combined in JSON-form data sent by the gateway node to the websocket server in the idcloudhost vps cloud via the websocket protocol. The following is a picture of 11 terminals on a vps cloud server.

```

Node 1
Soil Moisture 60.68%
Water Level 1.00cm
Data successfully sent to all clients

Node 2
Soil Moisture 62.80%
Water Level 1.20cm
Data successfully sent to all clients

Node 3
Soil Moisture 63.23%
Water Level 1.30cm
Data successfully sent to all clients

```

Figure 11. Receiving websocket data on the server.

Through this test, the results show that the client can receive water level and soil moisture data which have been combined in JSON-form data sent by the vps cloud server to the client via the websocket protocol. Here is a picture of 12 browsers display on the client.

1.Time	2.Node_Name	3.Soil_Moisture	4.Water_Level
2020-11-28T10:45:25	1	60.68%	1.00cm
2020-11-28T10:45:26	2	62.80%	1.20cm
2020-11-28T10:45:27	3	63.23%	1.30cm

Figure 12. Browser display on a client.

Testing the gateway node can send data packets and prove that the idcloudhost vps cloud server can receive water level and soil moisture data packets in the form of struct data sent by each sensor node via LoRa communication to be sent back to the websocket server in the idcloudhost vps cloud via the websocket protocol. Three data are sent, the first is the node name data, the second is the soil moisture data, and the last is the water level data. The data have also been entered into the cloud server database.

In addition, it also proves that the idcloudhost vps cloud server can send water level and soil moisture data packets in the form of JSON data that are sent to the client via the websocket protocol so that it can be displayed in table form on the web client application.

The sending of JSON data will go through the server IP number, which is 103.129.221.6 with port 9002. The use of port 9002 is one of the ports for the TCP or UDP transport layer. This conforms to the use of the websocket protocol, which also runs at the TCP transport layer.

Successful rate testing

The successful rate through this test, the success rate is in accordance with the tests conducted by the journal (Adin et al., 2019); it can be seen that the success of the packet can be sent safely and successfully influenced by the size and a small

distance used, which greatly affects the occurrence of the success rate. The following graph shows the average success rate based on distance.

Table 3. The quality category of the successful rate

Category	Successful Rate	Index
Excellent	100 - 98%	4
Good	97 - 86%	3
Medium	85 - 76%	2
Poor	<75%	1

According to the category table, figure 13 above illustrates that sensor node 1, sensor node 2, and sensor node 3 have an average total success of sending consecutive packages with each test for 30 min at 100 meters is 99.72% or in category excellent, at 200 meters it is 95.56% or in the good category, at 300 meters is 93.24% or in the good category, at 400 meters is 89.54% or in the good category, at 500 meters is 85.55% or with the medium category. Judging from the results of the graph above that package delivery is high. Meanwhile, having a total average decreases with increasing distance used. Based on the overall distance range, the total success rate of package delivery is 92.72% or in a good category.

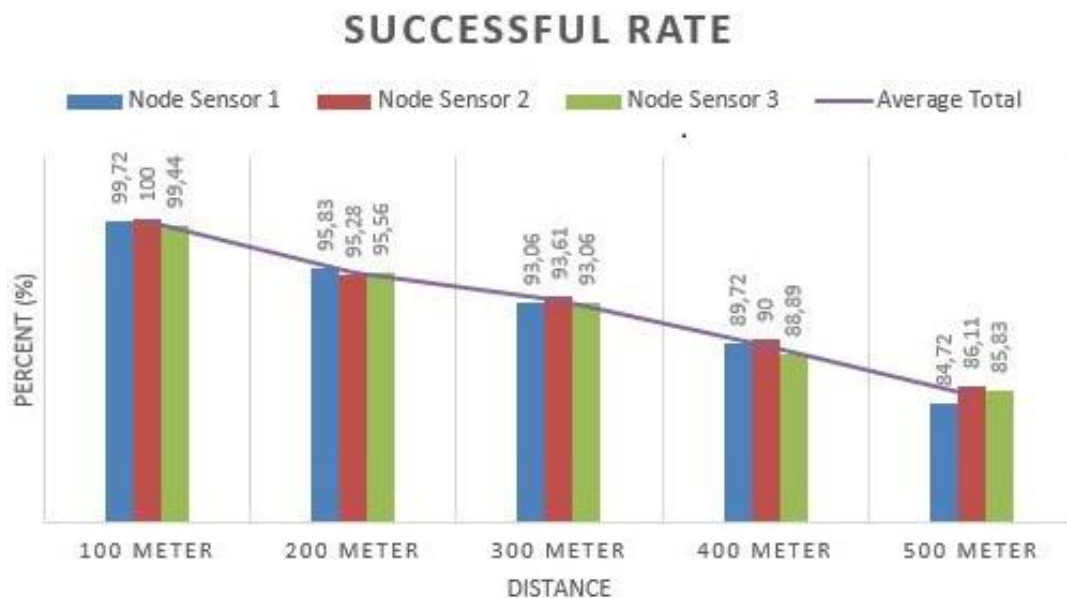


Figure 13. Successful rate graph.

Furthermore, Table 3 shows the successful rate categorization.

Packet loss testing

The next test, namely packet loss, through this test, the packet loss is in accordance with the tests conducted by the journal (Yanziah et al., 2020) can be seen that the unsuccessfulness of the packet influenced by the size and small distance used which greatly affects the occurrence of packet loss. The following graph shows the average packet loss based on distance.

Table 4. The quality category of the packet loss

Category	Packet Loss	Index
Excellent	0 - 2%	4
Good	3 - 14%	3
Medium	15 - 24%	2
Poor	>25%	1

According to the category table, figure 14 above illustrates that the sensor node 1, sensor node

2, and sensor node 3 have an average total unsuccessful packet delivery successively with each test for 30 min is at 100 meters is 0.28% or by category excellent, at 200 meters it is 4.44% or in the good category, at 300 meters it is 6.76% or in the good category, at 400 meters is 10.46% or in the good category, at 500 meters is 14.45% or with the medium category. Judging from the results of the graph above that packet delivery has a low packet loss or failure. Meanwhile, it has a total average that gets higher with the increasing distance used. Based on the overall distance range, it has an average total packet loss or unsuccessful delivery of packages, namely, 7.28% or in a good category.

RSSI signal testing

The next test, namely, the RSSI signal, can be seen that the signal strength of the packet can be transmitted safely and is successfully influenced by the distance used. The following graph shows the average RSSI signal based on distance.

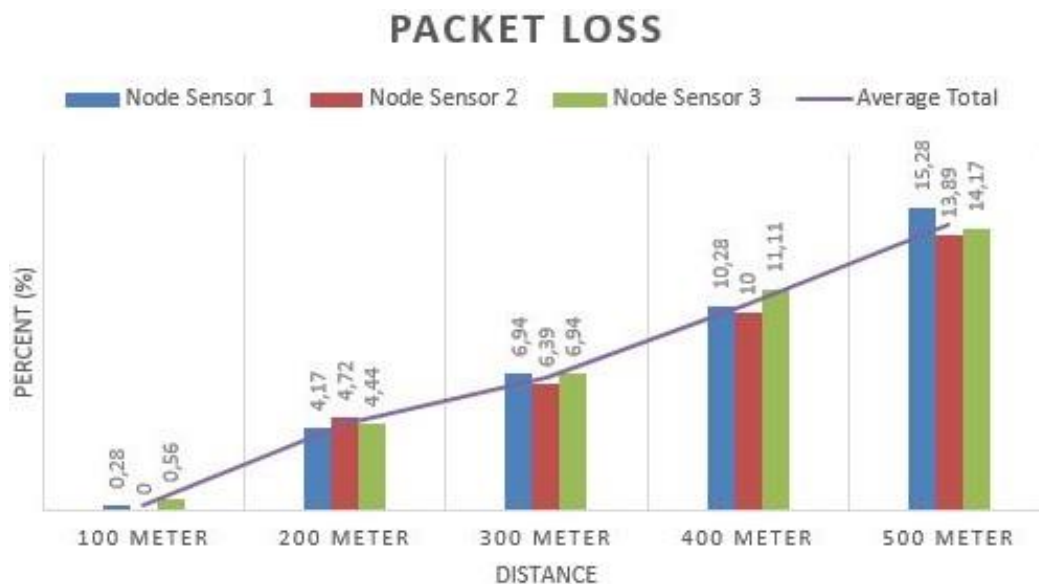


Figure 14. Packet loss graph.

Furthermore, Table 4 shows the packet loss categorization based on the TIPHON standard (Telecommunications and Internet Protocol Harmonization Over Network) (ETSI, 1999).

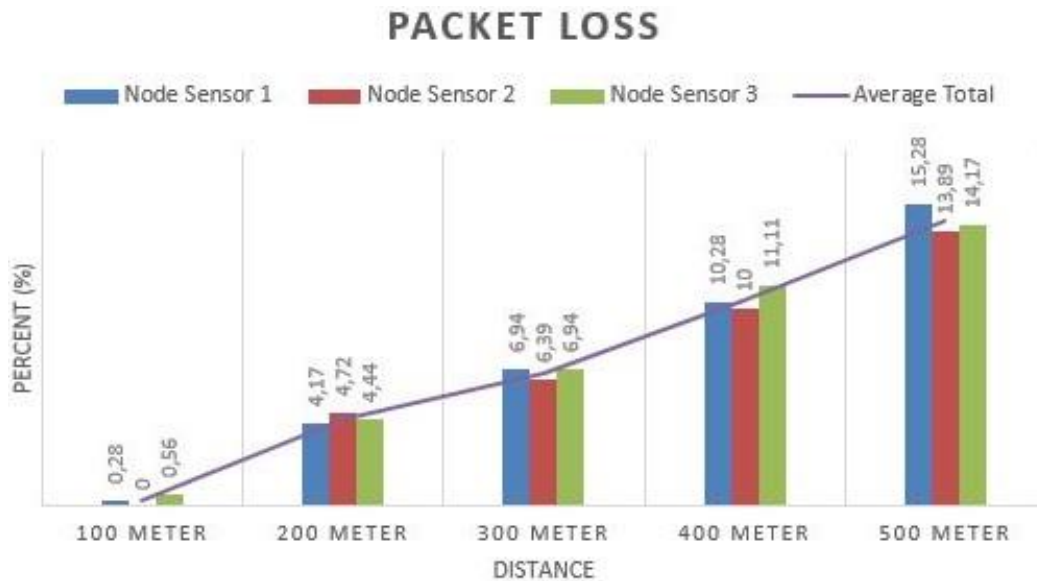


Figure 15. RSSI signal graph.

Furthermore, Table 4 shows the packet loss categorization based on the TIPHON standard (Telecommunications and Internet Protocol Harmonization Over Network) (ETSI, 1999).

Figure 15 above illustrates that the sensor node 1, sensor node 2, and sensor node 3 have an average total RSSI packet delivery signal in a row with each test for 30 min at 100 meters is -84.9 dBm, at 200 meters it is -95.15 dBm, at 300 meters it is -97.23 dBm, at 400 meters it is -99.96 dBm, at 500 meters it is -102.5 dBm. Judging from the results of the graph above that packet delivery has a good RSSI packet delivery signal. Meanwhile, it has a lower total average with the increasing distance used. Based on the overall distance range, the total RSSI signal for packet delivery is -95.95 dBm.

CONCLUSIONS

The reading of the sensor nodes equipped with the HC_SC04 ultrasonic sensor module and the MPX5700DP air pressure sensor which is equipped with a tensiometer, has succeeded in reading data. This is because the two sensors from each sensor node can acquire data well. The ultrasonic module itself can also provide good data accuracy with an average total error of 0.74% in detecting water level values that match the reality. The MPX5700DP pressure sensor module and Tensiometer can also provide good data accuracy with an average total error of 0% in detecting soil moisture values that match the reality.

LoRa data communication testing can also be said to be successful by sending struct data packets from the three sensor nodes to the gateway. Another test is sending through the websocket from the

gateway node to the cloud server so that data can be received and entered into the database server. In addition, the server can also send data to the client to be displayed on the client browser monitor.

LoRa communication performance is the successful rate found in packet delivery, which is a total average of 92.72% or in a good category. Packet loss with a total average of 7.28% or in a good category. RSSI signal (Received Signal Strength Indicator) with a total average of -95.95 dBm.

The advantage of this system is that monitoring the water condition of rice fields, especially with water level and soil moisture parameters, can be directly monitored online. So that farmers do not need to come directly to the farm to conduct monitoring.

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