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論　文　名	「PRACTICAL ATMOSPHERE CONTROL FOR PROLONGING SHELF LIFE AND PREVENTION OF SENESCENT SPOTTING IN BANANA FRUIT」 **（バナナ果実における品質保持と褐色斑点抑制のための ガス環境調整法）**
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Summary

INTRODUCTION

Banana (*Musa* spp.) is an economically important fruit in the tropical area with all year round production. The fruits quality depend on their own maturity and postharvest management. Banana ripe quickly after induction of ripening by ethylene. Browning spots, which are called senescent spotting or sugar spotting are observed just after the fruits are fully ripened and the taste of pulp reaches the maximum of acceptability. However, the phenomenon prevents consumers` choice when such banana decorate on the shelf of retail markets. Controlled atmosphere (CA) or modified atmosphere (MA) are beneficial for maturity control of many fruits. More practical modification of those effective treatments could useful for shipping and markets for fruits. Holding banana fruits in N_2 gas for short periods following ripening treatments can also delay ripening of banana. However, efficiacy of those treatments are not clear among former reports depending probably on the banana ripeness and actual low O_2 levels during application of the methods.

Our objectives were the studies of the potential gas treatment for prolonging shelf life of banana and the prevention of senescence spotting. We

also investigated initiation of senescent spotting by microscopic observation and change of phenolic compounds during developing spotting.

CHAPTER 1. STUDY OF SENESCENT SPOTTING OF BANANA (*Musa sapientum* L.) PEEL

There are two hypothesises on the initiation of the spots: one is the result of infection by fungus and the other is physiological disorder during senescence. So far the concrete mechanism of initiation of the spots is not clear.

1.1 Optical microscopic and scanning electron microscopic (SEM) observation

Less than two millimeter diameter of senescent spots of the surface peel (*Musa sapientum* L.) were observed by optical microscope and SEM. We found that the browning parts were about 0.25 mm depth, located within 7-8 layers of cells from surface. In horizontal view, the cells in browning spots showed 2-3 big brown particles in the cells. While no such particles were observed in the cells at the normal part. By SEM observation, the browning part showed sunken toward surrounding sound area like crater and wilted cells were observed in the crater. At the center of each sunken area, prominent stoma was observed. No hypha was observed on the senescent spotting area.

1.2 Sanitizing of peel by ethanol

Ethanol (20% or 50%) was treated to the surface of banana peels everyday for avoiding infection by microorganism during storage. There are no difference on initiation of browning between ethanol and non-ethanol treated banana. The senescent spotting of the two treatments started equally at day 7 and developed same extent thereafter.

These results in 1.1 and 1.2 section, showed that senescent spotting is a physiological disorder.

1.3 Browning and volatile chemicals

The yellow peels were separated from the pulp at day 6 after ethylene treatment when browning did not occur yet, and they were stored for successive 10 days at high humidity. The senescent spots did not occur on the

peels even after 10 days after peeling. We thought that some volatiles came out from pulp of banana, and went through the stomata and then denatured surrounding cells.

Volatile chemicals (5-100 μl) which reported earlier as aroma of banana were treated on the separated surface peel. The browning on the peel appeared in wider area, although typical senescent spots on the peel could not appear in any cases.

CHAPTER 2. PHENOLIC COMPOUND AND POLYPHENOL OXIDASE (PPO) ACTIVITY IN THE PEEL AND PULP

2.1 Sugar and dopamine levels during ripening

The total sugar content of banana pulp increased from 0.5% on day 0 to 8.7% on day 7 at 20℃. The dopamine, a catecholamine, of surface peel was about 500 mg per 100gFW on day 0 (green color fruit) and remained at that level until day 5 (yellow part). The brown part of senescent spotting on day 5 had lower content of dopamine (330 mg per 100gFW) than the yellow part. On day 7, the dopamine content at the senescent spotting showed further decrease. This indicated an inverse relationship between dopamine level and senescent spotting.

2.2 Polyphenol and PPO activity during ripening

The tyrosine and L-DOPA of the external peel were increased, while the dopamine was decreased during storage at 20℃. Dopamine was the majority among catecholamines. The catecholamines in pulp were low levels and not different during storage. Activities of PPO on external and internal peel as well as pulp were increased during ripening. These results imply that dopamine changes to browning pigments by PPO at the senescence stage of banana.

CHAPTER 3. MODIFIED ATMOSPHERE STORAGE IN POLYETHYLENE (PE) BAG AND POLYVINYL CHLORIDE (PVC) WRAPPING OF RIPENING-TREATED BANANA `SUCRIER`

3.1 MA storage in PE bags with or without ethylene absorbent (EA)

In the PE bag (0.03 mm thickness) at 20℃, the dopamine content of

surface peel kept higher level than in air (control). The sucrose levels of banana in PE and PE with EA were almost identical and lower than the level in the control. The concentrations of both carbon dioxide (CO_2) and ethylene (C_2H_4) in a PE bag were very high (about 27-32% CO_2 and 3 ppm C_2H_4, respectively). Slight fermented smell was observed, although PE packaging delayed ripening of the banana. The high level of CO_2 in the PE bag inhibited ripening. Therefore, the PE packaging seems unsuitable for MA storage at 20°C.

3.2 MA storage with PVC wrapping compared with PE bags

Banana in the PVC (0.01 mm thickness) wrapping at 20°C produced about 2.6% of CO_2 concentration on day 3, which increased to 4.7% on day 5 and then decreased to 3.0% from day 7 to day 13. The concentrations did not differ in wrappings with or without EA. The C_2H_4 concentration in PVC wrappings without EA was 0.7-1.0 ppm from day 3 to day 13, while in a wrapping with EA, the concentration was about 0.2 ppm from day 3 to day 6 and then decreased to 0.02-0.1 ppm from day 7 to day 13.

At day 7, the sucrose levels of banana in PVC with EA and the control were higher than that in PE with EA, although the glucose and fructose contents in all treatments were almost identical. Banana in PVC with EA had significantly higher dopamine contents than control banana and showed lower scores of senescence spotting.

There was a slower change of ripening in bananas held in PVC or PVC with EA at 20°C than in the control, and senescent spotting was also delayed. Shelf life was extended to 6-7 days, while the controls had a shelf life of only 3-4 days.

CHAPTER 4. NITROGEN (N_2)-GAS TREATMENT FOR SHORT TIME IN PE BAG FOR PROLONGING SHELF LIFE OF BANANA `SUCRIER`

N_2-gas (100%) treatment of banana in dynamic system for 48 h at 20°C, showed lower respiration rate about 40% than that in 21% oxygen (O_2)- gas treatment. Then the rate increased to normal air level after banana was transferred to the air flow condition after day 3. The treatment extended shelf life of banana to 5-6 days, while control had shelf life of 3-4 days. When the bananas were packed in PE bags with N_2 gas for 24 or 48 h at 25°C, the

O_2 concentrations in the PE bag were 0.98 and 3.41%, respectively, at the end of 24 and 48 h N_2-gas packaging. These bananas showed less senescent spots, high dopamine contents and 2 days longer shelf life compared to control.

A 24 h N_2-gas packing in PE bag and then PVC film wrapping extended the shelf life of banana to 9-11 days.

CONCLUSION

More practical controlling of surrounding N_2 gas was proposed on ripening of banana than conventional CA or MA storage. N_2-gas treatment in PE bags for short time (24 or 48 h) followed PVC wrapping made shelf life of banana twice and suppressed senescent spotting.

It was recognized that senescent spotting was physiological phenomenon during senescence and dopamine, a phenolic compound, decreased accompanying with development of senescent spotting.

審査結果の要旨

バナナは熱帯亜熱帯では生食にとどまらず、食糧としても重要な果実である。バナナはエチレン処理等によって追熟を開始すると老化までの過程が速やかに進み、黄化の後、果皮に褐色斑点が現れる。この時期には果肉の糖濃度が最高値に達し、香りも生成されて美味になり、この斑点はシュガースポットとも呼ばれている。しかし、小売店でこのような斑点が現れたバナナは消費者に嫌われる。そこで、小売店に配送されて追熟を開始した黄緑色バナナが黄化を経て褐色斑点を示すまでの期間をバナナの棚持ち期間と呼び、なるべくこの期間を延長することが良好な商品展示を行うために重要になってくる。果実の熟度を遅延する方法としては、低温で保存することや、ガス環境を厳密にコントロールする CA 貯蔵と、プラスチック袋に果実を入れて果実の呼吸とガスの膜透過性のバランスで好適なガス環境を作るＭＡ包装貯蔵がある。小売店の棚先で熟度を遅延するにはバナナが低温障害を示すことから低温貯蔵法は使えない。唯一ＭＡ包装が考えられるが、詳細には検討されていない。また一時的に窒素（N_2）ガスを処理して追熟の遅延を行うことも試みられているが、この処理の良し悪しは結論に達していない。

本研究においては、バナナ果実の棚持ち期間および褐色斑点出現を延長できる実際的な包装方法、N_2 ガス短期間処理方法およびその組み合わせを検討した。また褐色斑点そのものの発現の様相を光学および走査電子顕微鏡(SEM)で観察し、その発症起源を考察している。さらに褐色色素の由来をフェノール類の消長から示唆している。

その内容は以下のように要約される。

第 1 章では、褐色斑点を光学顕微鏡および SEM で観察することから始めた。バナナの褐色斑点が現れる原因として、真菌による感染と生理的障害が以前から言われている。現在のところ生理的障害が有力になっているが詳細は不明である。光学顕微鏡観察によって、褐色細胞は表皮の上層に限られているのが明らかにされた。ＳEMの観察では褐色斑点部分はクレーター様に陥没しており、中心には気孔があってその部分から褐色斑点が始まっている様子であった。また褐色斑点を示しているどの部分からも菌糸の存在は見られなかった。果皮をアルコール(20－50%)で毎日処理し、菌の付着を防止してみると、無処理(水)との間に差は無く、同時期に褐色斑点が現れた。これらの結果から、褐色斑点はバナナ追熟末期の生理的障害であることを強く示唆している。褐色斑点が気孔から始まっているようにうかがえたので、果肉の揮発性物質が関連しているのではないかと考え、剥皮した果皮にバナナ果肉で生成される化学物質の蒸気に曝したところ、幅広い褐変は起こるものの典型的な褐色斑点は現れなかった。

第 2 章において、褐色斑点発生時期と追熟中のフェノール類の含量、ポリフェノールオキシダーゼ（OPP）活性の変化との関係を見ている。主なフェノールはドパミンでその他 L-DOPA 等が少量存在していた。ドパミンは緑熟果から黄熟果にかけて高含量で

あったが褐色斑点が現れる頃に減少した。同一果実においても褐色斑点を示している果皮部分は低含量であった。またドパミンは果皮表層に多く見られ、褐色斑点との関係がうかがわれた。PPO 活性は同様に果皮表層で活性が高く、追熟中増加した。これらのことはドパミンが追熟後期に PPO の働きにより褐色色素に変化したことを示している。

第 3 章では、バナナを MA 包装したときの棚持ち期間および品質について、ポリエチレン（PE）袋(厚さ 0.03mm)とポリビニールクロライド(PVC) ラッピング（0.01mm）で比較した。20℃貯蔵において、PE 袋では内部の二酸化炭素（CO_2）濃度が高くなりすぎて(約 30%)発酵臭と追熟障害を起こし、包装としては適切ではなかったが、PVC でラッピングする方法は内部のガス濃度(CO_2, 3-4%)も適当で、無包装区(3－4 日)に比べ、棚持ち期間を延長することができた(6－７日)。

第 4 章では、100%窒素ガス(N_2)短時間処理の棚持ち性および品質に与える影響を見ている。ここで述べる N_2 ガス処理は実用面から考えて大型の PE 袋(厚さ 0.05mm)にバナナ果実をいれ、100%N_2 ガス処理を 24 時間または 48 時間行っている。この方法では、処理完了前の酸素（O_2）濃度はそれぞれ 0.98 または 3.41%であった。この処理も前章の PVC ラッピングと同程度に棚持ち期間を延長した。上述の方法を組み合わせて、100%N_2 ガス処理を 24 時間行い、その後 PVC ラッピングすることにより棚持ち期間を 9－11 日に延長することができた。

本論文は、バナナの流通小売上の棚持ち期間の大幅な改善方法を提示するとともに、バナナの貯蔵後期に現れる褐色斑点の顕微鏡による観察やフェノール含量、PPO 活性の変化から、褐色斑点の発現機構も示唆している。この成果は実際的な流通上の問題点を改善したことにとどまらず抜本的な果実の褐変現象の解明に役立つデーターを多く含んでいる。よって、学力確認の結果とあわせて、博士(学術）の学位を授与することを適当と認める。