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ダチョウ卵白主要タンパク質の消化性に関する研究

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鶏卵のもつ食生活上の意義は言うまでもないが、流通量の少ない他の食卵成分における栄養学的・食品学的な意義に関する研究はほとんどない。しかしながら、分類学的に離れた鳥卵の場合には、鳥卵構成タンパク質の構造も大きく異なると推察され、それに伴いタンパク質の機能（特性）にも影響があると思われる。とくに、大きな健康問題である卵アレルギーに関連して、鳥卵タンパク質の種類、化学構造や消化性は、食の安全を保つ上で基本的な知見と思われる。そこで本研究では、ニワトリ（Chi）と分類学的に離れ、近年新たな食材にもなっているダチョウ（Ost）卵に関して、主要タンパク質のオボアルブミン（OVA）およびオボムコイド（OVM）を分離・同定し、それらの消化性を鶏卵と比較しながら明らかにした。また、その間、OstOVMの構造・構造変化や分解ペプチド追跡のツールとして、モノクローナル抗体（MAb）の作製も行った。

第1章では、Ost卵白のポリペプチド構成等を明らかにしてChiと比較した。次いで、抗ChiOVAおよびOVM抗体により、OstOVAおよびOVMを免疫学的に同定した。さらに、精製方法を確立してOstOVAおよびOVMを精製した。

第2章では、特異抗血清およびOstOVM特異的MAbを作製した。ハイブリド-マ65株のうち5クローンから得たMAbは、すべて不連続型エピトープ結合抗体の特徴をもち、糖鎖への特異性はほぼなかったが、各種鳥卵OVMとの反応性から、5種のMAbとも異なる特異性をもっていた。また、OstOVMの加熱処理により結合性が増すMAbと、構造変化には感受性が低いMAbの二つのタイプに分けられた。

第3章では、人工胃液（SGF）および人工腸液（SIF）を用いてOstおよびChiのOVAとOVMの消化性を比較した。加熱処理タンパク質の消化性やSGF/SIF連続消化の効果についても検討を行った。その結果、OstOVA、OVMともにChiより消化性が劣っていたが、適度な加熱により改善されることがわかった。このことは抗原性ペプチドの残存やアレルギー反応惹起という点で、ダチョウ卵消費にあたっては十分な加熱等の注意が必要と思われた。なお、第2章で作製した5種のMAbのうち3種のMAbにより、OstOVMから生成する抗原性ペプチドが高感度で検出でき、今後、これらのMAbを利用して、抗原性ペプチドの分離、追跡等が可能になると思われた。

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Digestibility of the major proteins in ostrich egg white

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Although chicken eggs have long been recognized as an excellent dietary resource, scientific information is limited on the nutritional and functional properties of other avian eggs that are rare in the market. The egg proteins of birds genetically distant from chickens might have different structure and functions from those of chicken-orthologous proteins and thus knowledge about the composition of such egg proteins, their chemical structure and the digestibility is quite fundamental for food safety, especially for egg- allergic patients. In the present study, ostrich (Ost) egg were focused on, since Ost is genetically distant from the chicken and the consumption of Ost eggs is now growing. The major proteins of Ost egg, ovalbumin (OVA) and ovomucoid (OVM) were isolated from Ost egg white and their digestibility was investigated. Moreover, the hybridoma-producing monoclonal antibodies (MAb) specific to OstOVM were established in order to obtain the tools for detecting OstOVM, its structural changes and derived peptides.

In Chapter 1, the polypeptide composition of Ost egg white proteins was compared with that of chicken egg proteins. Then, Ost OVA and OVM were immunologically identified. After establishing the fractionation methods, Ost OVA and OVM were purified.

In Chapter 2, murine antisera specific to Ost OVA and OVM were raised. Sixty five hybridomas were established and 5 MAbs were obtained in ascites fluid. The epitopes recognized by the MAbs were suggested to be a discontinuous type and apart from the glycosylation sites. The MAbs were distinct in specificities judging from the extent of binding to various OVMs from 9 avian species other than ostrich. They were also classified into two groups based on the specificity to heat-treated OstOVM: one was more specific to heated OVM and another was indifferent to the heat treatment.

In Chapter 3, the digestibility of OVA and OVM from Ost and Chi by simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) was compared. The effects of heat treatment of the proteins and SGF digestion followed by SIF digestion were also examined. Consequently, the digestibility of Ost proteins was inferior to that of Chi proteins but improved by adequate heat treatment. This suggests that appropriate care such as sufficient heat treatment should be taken in the consumption of Ost eggs in order to lower the absorption of antigenic peptides causative of allergy. In addition, antigenic peptides derived from OstOVM were sensitively detected with 3 MAbs out of 5 MAbs obtained in Chapter 2, showing the availability of the MAbs in the isolation and tracing of the antigenic peptides.