TRENDS OF BIOTECHNOLOGY RESEARCH IN ANIMAL PRODUCTION IN VIETNAM

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ABSTRACT

Since 1994, biotechnology has been developed in various scopes of animal production in Vietnam. Detection of genes relating to production performance is the target in pigs whereas *in vitro* production of embryo is dominant in cattle. Studies in feed additive mainly focus on bacterial enzymes. Regarding to animal health, molecular techniques are widely applied for detection of important diseases in domestic animals and poultry. So far, recombinant proteins used in diagnosis and production of sub-unit vaccines are just in sequencing to identify the structures. Moreover, application of biotechnology in drug production and environment protection is still limited.

Key words: biotechnology, research, animal production, Vietnam

INTRODUCTION

The population of different species of animals has increased at the rate of 5-10% in Vietnam. In 2005, there were 5.4 million cattle including 104,000 dairy cows that covered 22% of the milk demands, 2.92 million buffalo, 27.4 million pigs from which 36.2 million pork were slaughtered, 1.31 million goat and sheep, 160 million chicken and 60 million duck. The animal production is going to the trend of industrialization and specialization, and its contribution to agriculture GDP averages 23%. Meat is produced mainly for local consumption, and some carcasses of feeder pigs are exported to Hong Kong. The limitations of animal production are a lack of large herds of genetically improved breeders, poor quality of feed ingredients, disease spread and inefficient control of environment, all of which may be partly solved through biotechnology development.

Government Decision of 1994 indicates five focal points of biotechnology development till 2010, including biotechnology in agriculture and animal production, public health, protection of environment and natural resources, other industries, and establishment of biotechnological system. In animal production, the main approaches of biotechnology research go towards the improvement of animal breeds involving gene detection and gene conservation; assistant reproduction techniques; production of additive feed such as probiotic, vitamins and amino acid; disease detection; production of recombinant proteins specific for diagnostic kits and vaccines; finding some microorganisms useful for waste treatment.

APPROACHES OF RESEARCH

Gene detection and genetic improvement

In swine

Pork is a major source of protein (68%) in meals of Vietnameses; therefore, the detection of genes controlling the production performance has been focused.

- The gene mostly studied is *halothane gene* (ryanodine 1 receptor gene) that has two alleles. Its association with growth rate, reproductive performance, carcasses quality and meat quality has been defined (Nguyen Ngoc Tuan and Tran Thi Dan, 2001; Nguyen Van Cuong et al, 2002). However, mating the pigs based on the presence of the halothane genotypes is still limited.

- Two genes involving the litter size - *estrogen receptor gene* and *prolactin receptor gene* are the second interest in pigs of local as well as exotic breeds. Regarding to estrogen receptor gene, BB-genotype sows perform a larger number of piglets than AA-genotype ones by 5-15% (Le Thi Thuy et al, 2002) but there is not relationship between the genotypes and semen quality (Tran Thi Dan et al, 2005). In addition, the gene coding for *gonadotropin releasing hormone receptor* (GnRHR) in one native breed and Yorkshire is sequenced (Nguyen Dang Ton et al, 2004), which may be useful for improving the reproductive performance of pig because GnRH regulates the secretion of two pituitary hormones controlling reproduction of mammalian animals.

- The polymorphisms of three other genes - *pituitary-specific transcription factor-1 gene*, *myogenin gene* and *heart fatty acid BP gene*, associating with growth rate, muscle differentiation and muscular fat, respectively, are found in some native breeds of pigs (Pham Thu Thuy et al, 2003; Nguyen Van Anh et al, 2005; Nguyen Thu Thuy et al, 2005).

Cattle

In cattle, less work on gene detection has been done than *in vitro* production of embryo. Markers relating to κ casein and β lactoglobulin of milk are used to determine some genotypes favoring the ability of milk production.

In chicken

Method of directly injecting DNA into the pronuclear of chicken embryo is applied to produce a chimera chicken from one local breed and one Chinese breed named Luong Phuong. The performance of the chimera has not yet reported.

Marker is also used to find the presence of growth-hormone gene in poultry of local breeds. However, the association between the gene and production performance was not consistent.

Embryo transfer and sex determination

In vitro maturation of oocytes and *in vitro* production of embryo have been studied in cattle (Nguyen Quoc Dat et al, 2003; Bui Xuan Nguyen, 2004; Tran Thi Dan et al, 2005), pig (Huynh Thi Le Duyen, 2003) and dog (Tran Thi Dan et al, 2005). The protocols of sex determination for cattle and pigs are also achieved. A number of cattle produced from *in vitro* embryo were born; however, the commercial production has not yet established. In addition, cryobanking for embryo and tissues has been set up under the national project.

Feed additive

Strains of Bacillus, especially *Bacillus subtilis*, are intensively studied to explore their biomass (To Minh Chau et al, 2005) as probiotic, and to use their enzymes for enhancement of digestion in animals and human. Protease is an important industrial enzyme, accounting for 60% of the total enzyme market; hence, protease of *Bacillus subtilis* is purified and produced (Do Thi Bich Thuy and Tran Thi Xo, 2004). The nucleotide sequence of the 16S-RNA gene of the strain Bacillus HA401 that produces alkaline alpha amylase is also demonstrated (Tang Thi Chinh, 2006).

Another enzyme – phytase, that enhances phosphorus digestibility, is produced from *Aspergillus niger* (Tran Thi Tuyet et al, 2004). The enzyme contributes to improving the feed conversion ratio and declining the environment pollution due to phosphorus.

S-adenosyl-L-methionine (SAM) is a natural substance produced from adenosine triphosphate and methionine. It works as a source of nutrient for many functions in body. The product is successfully extracted from *Saccharomyces cerevisiae* biomass (Tran Thi Huong et al, 2005).

Veterinary medicine

PCR and cellular culture in diagnosis

PCR is widely used to diagnose the important bacteria and viruses in domestic animals and fowl. Biotechnology approaches in studying parasites has been very limited. The major microorganisms examined:

- Virulent types of *E. coli* (Nguyen Ngoc Tuan et al, 2005) and *Campylobacter* (Vo Ngoc Bao et al, 2006) associating with diarrhea in animals and food-born disease in human

- *Mycoplasma* infection in pig (Nguyen Thi Phuoc Ninh et al, 2006) and chicken (Nhu Van Thu, 2006), that opens the opportunity for secondary infections of lungs.

- Foot and mouth disease virus causing serious problems to health and economic value of cattle, buffalo and pig (To Long Thanh et al, 2004)

- Different types of hog cholera virus that infect brain and many systems of the body including gastrointestinal tract in pigs (Nguyen Thi Thu Hong et al, 2003)

- Viruses of enzootic diseases relating to loss of production performance in pigs, including porcine reproductive and respiratory syndrome virus (Tran Thi Bich Lien and Tran Thi Dan, 2003), Japanese encephalitis virus carried by pigs and transmitted by mosquito (Ho Thi Viet Thu et al, 2006) and porcine circovirus type 2 causing the wasting syndrome of young pigs (Lam Thi Thu Huong and Duong Chi Mai, 2006).

- In pigs, a new bacterium genus *Arcobacter* that together with Campylobacter has been found in faces and dead fetuses (Hoa et al, 2006), which may be a source of food-born disease

- Gumboro virus causing immunodeficiency in chicken (Quyen Dinh Thi et al, 2004), and subsequently inducing the second infections

- Bird flu virus – one of the current agents of lung infection in fowl and human (Nguyen Tien Dung et al, 2004).

Production of recombinant antigen

Besides the production of the traditional vaccines for a long time to control some dangerous diseases caused by hog cholera virus, Newcastle disease virus, *Pasteurella multocida*, the recombinant antigens specific for diagnosis or sub-unit vaccine are also developed.

- VT2e toxin produced by *E. coli* is extracted and used as sub-unit vaccine to prevent edema disease of young pigs (Nguyen Ngoc Hai and Milon, 2005).

- *Salmonella enteriditis* is one of Salmonella types causing food-born diseases, which is carried by domestic animals. Bacteraemia and diarrhea are

the common symptoms. The *gm3* gene encoding the flagella antigen (H: g,m) of the bacteria is cloned and expressed in *E. coli* (Nguyen Hong Thanh et al, 2004).

- Rabies virus is still threatened to human and animals, especially dogs. Glycoprotein is the most important antigen for immunization against the virus; therefore, gene coding for the glycoprotein is sequenced (Chu Hoang Ha et al, 2005).

- Envelope protein of the Japanese encephalitis virus is expressed through recombinant baculovirus (Nguyen Thanh Thuy Nhien et al, 2005).

- Virulent gene *vp2* and its antigen are isolated from Gumboro virus associating with immunodeficiency in chicken (Quyen Dinh Thi et al, 2004). The result opens the opportunity to produce vector DNA vaccine for controlling one of the dangerous infectious diseases in chicken.

Antibiotic production and antibiotic resistance

The production of vancomycin – a glycopeptide antibiotic, by fermentation is of interest for the importance of the product against pathogens resistant to many popular antibacterial agents. *Streptomyces orientalis* 4912 is studied with respect to the biology and cultivation characteristics, and vancomycin production (Nguyen Phuong Nhue et al, 2004).

In contrast to antibiotic production, the genetic resistance of bacteria to antibiotic has been indicated. An et al (2006) has found the class 1 integrons and the characteristic of resistance gene cassettes integrated in these integrons of non-typhoid *Salmonella* isolated from cattle, pig and chicken.

Environment protection in animal production

Biogas and compost are the main solutions for animal waste treatment. Bacteria packages facilitate the fermentation are commercially available.

To solve the pollution due to aromatic compounds that are toxic to animals and human, and difficult to be degraded, bacteria is one of the effective measures. *Rhodopseudomonas palustris* can grow with its versatile metabolic modes, utilizes and degrades various aromatic compounds. The gene relating to aromatic compound metabolism is detected in *Rhodopseudomonas palustris* using recombinant *E. coli* (Van Thi Nhu Ngoc, 2005).

Denaturing gradient gel electrophoresis (DGGE) is applied to study the mutation and diversity of microorganisms, especially the microbial community in toxic chemical - contaminated areas (Nguyen Thanh Thuy et al, 2004).

Additional notes on biotechnology in aquaculture

Three main areas of biotechnology research in aquaculture are (1) finding polymorphism of fish and shrimp using markers (Nguyen Van Cuong et al, 2004; Nguyen Thi Thao et al, 2004), and (2) disease detection, especially white spot syndrome caused by virus (Phan Thanh Phuong et al, 2003; Nguyen Hoang Nam Kha et al, 2006; Tran Nhat Minh et al, 2006), and (3) disease treatment (Ly Thi Thanh Loan et al, 2006).

CONCLUSION

Biotechnology has been applied in various aspects in animal production in Vietnam. However, there is still a gap between the lab outputs and the production of commercial packages.

REFERENCES

AN T.T. VO, VAN DUIJKEREN E., FLUIT C.A. and GAASTRA W., 2006. Characterization of resistance genes associated with class I integrons in non-typhoid *Salmonella*. Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

BUI XUAN NGUYEN, 2004. Development of embryo biotechnology at Vietnam Academic for Science and Technology. *Proceedings of the First Workshop of the Asian Reproductive Biotechnology Society*, Nong Lam University HCMC, Vietnam, 12-14 April, 2004.

CHU HOANG HA, NGUYEN THI THU TRANG, LE THI HONG NGOC, LE TRAN BINH and NONG VAN HAI, 2005. Sequence analysis of gene coding nucleoprotein N of rabies virus. *J. Biotechnology* (in Vietnamese) 3 (3):339-346.

DO THI BICH THUY and TRAN THI XO, 2004. Production, purification and application of protease from *Bacillus subtilis*. *J. Agri. Sci. and Technol.* (in Vietnamese) 4/2004: 37-41.

HO THI VIET THU, LUU LE LOAN, HUYNH NGOC TRANG, DU VU KHANH, BUI THI MY NHIEN, NGUYEN VAN AN, NGUYEN TRI NHAN and TRAN DINH TU, 2006. Components of mosquitoes transmitting Japanese encephalitis virus and effects of natural factors on mosquito density in Can Tho City. *J. Vet. Sci. and Technol*. (in Vietnamese) XIII (3): 60-66.

HOA T.K. HO, LIPMAN J.A.L., VAN DER GRAAF-VAN BLOIS L., VAN BERGEN M., TUAN N.N. and GAASTRA W., 2006. Application of molecular approaches in study of *Arcobacter* spp. prevalence in pigs and their transmission from infected sows to newborn piglets. Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

HUYNH THI LE DUYEN, 2003. Application of in vitro fertilization and PCR to determine sex of pig. MSc. thesis, National University HCMC.

LAM THI THU HUONG and DUONG CHI MAI, 2006. Detection of porcine circovirus from lesions of postweaning pig with wasting disease at some farms in HCMC and some adjacent provinces. Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

LE THI THUY, PHAM DOAN LAN, NGUYEN VAN HAU, TRAN THU THUY, LUU QUANG MINH and NGUYEN DANG VANG, 2002. Application of molecular genetics to determine gene relating to reproductive traits of pigs in Vietnam. *J. Animal Husbandry* (in Vietnamese) 8: 7-9.

LY THI THANH LOAN, VO HONG PHUONG, THIEN PHUC THANH and NGUYEN THI HUYEN, 2006b. Experimental screening some pharmaceutical chemistry compounds affecting *Achlya* sp. isolated from catfish eggs. Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

NGUYEN DANG TON, PHAM THU THUY, PHAM THI VAN HOA, LE THI THU HIEN, LE THI THUY, NGUYEN DANG VANG, NONG VAN HAI, 2004. Polymorphism of the gene coding for gonadotropin releasing hormone receptor (GnRHR) in 'I' and Yorkshire pigs. *J. Biotechnology* (in Vietnamese) 2 (4): 431-438.

NGUYEN HOANG NAM KHA, SARTER S., LEGARVE T., MONTER D., 2006. Characterization of bacteria isolated from farmed catfish coming from Vietnam and study of their antibiotic resistance. Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

NGUYEN HONG THANH, TRAN NGOC TAN, DO THI HUYEN, TO LONG THANH and TRUONG NAM HAI, 2004. Expression of *gm3* gene encoding flagella H: g,m antigen of *Salmonella enteriditis*. *J. Biotechnology* (in Vietnamese) 2 (4): 415-420.

NGUYEN NGOC HAI and MILON A., 2005. Recombinant *Escherichia coli* expressing the fusion protein B-subunit of the VT2e. J. *Agri. Sci. and Technol.* (in Vietnamese) 1: 14-19.

NGUYEN NGOC TUAN and TRAN THI DAN, 2001. Effect of stress-inducing gene (halothane gene) on growth rate, carcass quality and reproductive performance in pigs. *J. Animal Husbandry* (in Vietnamese) 7: 13-14.

NGUYEN NGOC TUAN, BUI THI THU TRANG, LE THI MAI KHANH and TRAN THI DAN, 2005. Detection of some virulent genes of *E. coli* in dung of cattle and pigs using multiplex PCR. *Agri. Sci. and Technol.* (in Vietnamese) 1/2005: 20-25.

NGUYEN PHUONG NHUE, NGUYEN VAN HIEU and LE GIA HY, 2004. Biological characteristics of *Streptomyces orientalis* 4912 producing vancomycin. *J. Biotechnology* (in Vietnamese) 2 (4):511-516.

NGUYEN QUOC DAT, NGUYEN VAN TIM and LE XUAN CUONG, 2003. Application of biotechnology in production and transfer of embryo to rapid increase the number of dairy cattle of high milk yield in HCMC. Report.

NGUYEN THANH THUY, LA THI THANH PHUONG and DANG THI CAM HA, 2004. Using denaturing gradient gel electrophoresis for detection of microbial community in toxic chemicals – contsminated soil. *J. Biotechnology* (in Vietnamese) 2 (3): 381-388.

NGUYEN THANH THUY NHIEN, KAEKO KAMEI, HAJIME MORI and TRAN LINH THUOC, 2005. Construction of a recombinant baculovirus encoding Japanese encephalitis virus envelope protein. *J. Biotechnology* (in Vietnamese 3 (1): 29-38.

NGUYEN THI PHUOC NINH, DO TIEN DUY, NGUYEN TAT TOAN, NGUYEN NGOC TUAN and TRAN THI DAN, 2006. Isolation of *Mycoplasma hyopneumoniae* and some bacteria relating to pneumonia diseases in pig lungs. *J. Vet. Sci. and Technol*. (in Vietnamese) XIII (3): 5-11.

NGUYEN THI THAO, QUYEN DINH THI, DAO THI TUYET, LE THI THU GIANG and PHAM ANH TUAN, 2004. Genetic variation of three populations of cultured Vietnam black tiger shrimp (*Penaeus monidon*) by microsatellite. *J. Biotechnology* (in Vietnamese) 2 (3): 315-324.

NGUYEN THI THU HONG, NGUYEN TIEN HA, NGUYEN TIEN TRUNG, NGUYEN VAN DUNG and MORRISSY J.C., 2003. Passive immunity of piglets against hog cholera virus in Southern Vietnam. *J. Vet. Sci. and Technol*. (in Vietnamese) X (4): 13-20.

NGUYEN THU THUY, NGUYEN THI DIEU THUY, NGUYEN KIM DO and NGUYEN VAN CUONG, 2005. Genetic variation of heart-fatty acidBP gene in five breeds of Vietnam *J. Biotechnology* (in Vietnamese) 3 (3): 303-309.

NGUYEN TIEN DUNG, PEIRIS M., WEBSTER R., DAO THANH VAN, BUI NGOC ANH, NGUYEN THE VINH, INUI K., BUI NGHIA VUONG, NGUYEN VIET KHONG and NGO THANH LONG, 2004. Source of bird flu virus H5N1 in Vietnam during 2003-2004. *J. Vet. Sci. and Technol*. (in Vietnamese) XI (3): 6-14.

NGUYEN VAN CUONG, NGUYEN THI DIEU THUY, NGUYEN THU THUY, LE THI PHUONG THAO, NGUYEN KIM DO, NGUYEN VAN DUC, NGUYEN DANG VANG, 2002. Frequency of genotype RYR-1 in pigs of some breeds using PCR-RFLP. *J. Genetics Application* (in Vietnamese) 4: 26-31.

NGUYEN VAN CUONG, NGUYEN THU THUY, NGUYEN THI DIEU THUY, DANG THANH HA and PHAM ANH TUAN, 2004. Development of AFLP markers for growth in catfish (*Pangasius hypophthalmus*). *J. Biotechnology* (in Vietnamese) 2 (3): 307-313.

NGUYEN VAN ANH, NGUYEN THI DIEU THUY, NGUYEN VAN CUONG and NGUYEN KIM DO, 2005. Genetic variation of myogenin gene in Mong Cai pig. *J. Biotechnology* (in Vietnamese) 3 (3): 311-317.

NHU VAN THU, MAILLARD C.J., CU HUU PHU, PHAN THANH PHUONG, LE MINH SAT, TRUONG QUANG LAM, 2006. Some differences in sequences of 16S rRNA gene of *Mycoplasma* strains isolated from poultry. *J. Vet. Sci. and Technol*. (in Vietnamese) XIII (1): 26-32.

PHAM THU THUY, LE THI THUY, CAO XUAN HIEU, NGUYEN DANG TON, LE THI THU HIEN, NGUYEN DANG VANG, NONG VAN HAI, 2003. Sequencing PIT-1 gene in pigs of breeds Landrace, Ba Xuyen and My Van. *J. Biotechnology* (in Vietnamese) 1 (3): 317-324.

PHAN THANH PHUONG, PHAM CONG HOAT et al., 2003. Application of molecular biotechnology in diagnosing and preventing white spot disease in shrimp. *J. Vet. Sci. and Technol*. (in Vietnamese) X (4): 61-66.

QUYEN DINH THI, LE THI THU GIANG, TRAN THI BICH HONG, LE THANH HOA, PHAM VIET CUONG, 2004. Cloning and sequence analysis of the *vp2* gene from three Gumboro virus strains isolated in Vietnam. *J. Biotechnology* (in Vietnamese) 2 (4): 439-449.

TANG THI CHINH, 2006. Taxonomy of alkalophilic Bacillus strains by analysing the nucleotide sequence of the 16S-RNA gene. *J. Biology* (in Vietnamese) 28 (1): 63-67.

TO LONG THANH, TRUONG VAN DUNG, LE VAN KHANG, DINH DUY KHANG, DUONG HONG QUAN, 2004. Diagnosing types of FMD virus using RT-PCR. *J. Vet. Sci. and Technol*. (in Vietnamese) XI (3): 15-21.

TO MINH CHAU, LAM THI THU HUONG and NGUYEN DUC DUY ANH, 2005. Determining the optimal medium of culture to harvest biomass and enzyme of *Bacillus subtilis*, *Lactobacillus acidophilus,* and production of primary package. *J. Agri. Sci. and Technol.* (in Vietnamese) 2-3: 16-20.

TRAN NHAT MINH, TRAN THI THANH XUAN, LE PHUC CHIEN, NGUYEN THI THU HANG, NGUYEN THI HONG VAN, PHAN THI CAM LINH, VAN THI HANH, and NGUYEN QUOC BINH, 2006. Detecting a new TSV-like virus casuing disease on tiger shrimp (*Penaeus monodon*). Proceedings of Biotechnology Workshop, Nong Lam University HCMC, Vietnam, in press.

TRAN THI BICH LIEN and TRAN THI DAN, 2003. Prevalence of PRRS and some clinical signs of reproductive and respiratory syndrome in pigs at one farm. *J. Vet. Sci. and Technol*. (in Vietnamese) X (4): 89-91.

TRAN THI DAN, NGUYEN NGOC TUAN, LE THI THANH HUE, 2005. Variation of swine semen quality in different genotypes of estrogen receptor gene and concentration of reproduction hormones in blood. *J. Agri. Sci. and Technol.* (in Vietnamese) 2-3: 37-39.

TRAN THI DAN, QUACH TUYET ANH and NGUYEN BACH THAO VY, 2005. Application of protocol for in vitro maturation of canine ooytes. *J. Agri. Sci. and Technol.* (in Vietnamese) 2-3/2005: 34-36.

TRAN THI DAN, QUACH TUYET ANH and NGUYEN VAN UT, 2005. Determination of sex in cattle of three breeds using multiplex PCR. *J. Agri. Sci. and Technol.* (in Vietnamese) 2-3/2005: 30-33.

TRAN THI HUONG, PHAM THUY HONG, NGUYEN THI HOAI TRAM and TRUONG NAM HAI, 2005. Extraction and purification of S-adenosyl-L-methionine from *Saccharomyces cerevisiae* IFO 2346. *J. Biotechnology* (in Vietnamese) 3 (1): 115-121.

TRAN THI TUYET, NGUYEN DUY LONG and HOANG QUOC KHANH, 2004. Production of phytase by *Aspergillus niger* NRRL 363. *J. Agri. Sci. and Technol.* (in Vietnamese) 4:28-32.

VAN THI NHU NGOC, DINH THI THU HANG, DO THI HUYEN, TRAN VAN NHI and TRUONG NAM HAI, 2005. Expression of BADA gene coding benzoat coenzyme A ligase from *Rhodopseudomonas palustris* in *E. coli*. *J. Biotechnology* (in Vietnamese) 3 (3): 319-324.

VO NGOC BAO, NGUYEN NGOC DIEN and FRIES R., 2006. Prevalence of *Campylobacter* on chicken carcass at slaughter houses in Ho Chi Minh City. *J. Vet. Sci. and Technol*. (in Vietnamese) XIII (1): 33-37.