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SPECIAL FEATURE

Papers dedicated to Professor Motoo KIMURA on the occasion of his 70th birthday

A note on the neutralism

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ABSTRACT

This note is about the neutralism first proposed by Dr. Motoo Kimura. It consists of two sections. The first section describes how one person became a neutralist. The second section is a preliminary report on the historical research of neutralism based on questionnaire sent to members of the Genetics Society of Japan.

1. INTRODUCTION

This is not an article reporting results of original research in genetics and related subjects nor a review paper. I hope that this note, written by a devoted neutralist, might still be appropriate as a paper dedicated to Professor Motoo Kimura on the occasion of his seventieth birthday.

This note is divided into two sections. The first section titled "Becoming a devoted neutralist" is a personal memoir of myself. The second section is about the project jointly carried out with Dr. William Provine, on the history of neutralism. A preliminary result of the questionnaire sent to members of Genetics Society of Japan is described.

2. BECOMING A DEVOTED NEUTRALIST

When I was a freshman of biology major about 20 years ago, I was not sure which subject to study in the future. Although I got interested in genetics and evolution to some extent, there were many other exciting fields of biology. Thus I took various special seminars as long as there was vacant lecture time not occupied by obligatory courses. Reading of classic biology papers organized by late Dr. Shinsaku Yamakawa was one of those seminars, and I read papers by A. Weismann, G. H. Hardy, and by Th. Dobzhansky among others. I immediately started to respect the theory of Weismann (continuity of the germ-plasm), but the papers by the other two were not so interesting for me.

I decided to choose physical anthropology for my major, and got acquainted with junior and senior students of the department of anthropology while I was a sophomore. One day, when I talked with a junior student, late Mr. Shu Kamata, he mentioned the neutral theory. Because I did not know even the name of the theory, he chuckled and showed me a copy of an article appeared in *Kagaku* (Kimura, 1976). To read that article was an assignment to junior students given by Dr. Keiichi Omoto. I read that, and somehow I immediately fell in love with the neutral theory.

There were at least three aspects in my affection toward the neutralism; (1) clear biochemical basis on the proposal of the theory, (2) extraction of simple and beautiful formulations through rigorous mathematical treatments, and (3) possibility of deriving a new view of life. Aspects (1) and (2) are unnecessary to explain, while some people may be puzzled what aspect (3) means. The idea that even a mutant with no selective advantage can be used in evolution is, in my mind, connected to a certain world view. I will come back to this point later.

Until that time I had some knowledge of evolution through reading introductory books. However, I was not satisfied with the explanation about the mechanism of evolution based on Neo-Darwinism. For example, a complicated character was considered to be attained through successive fixation of advantageous mutations. But is it possible to always have an increment of fitness by a new mutation to be fixed in a population? One alternative explanation was that of Imanishi (e.g., Imanishi, 1976), who advocated the existence of mysterious force in mutation. Although I shared a doubt against the Neo-Darwinian explanation with him, Imanishi's "idea" was not able to become science, and I neglected his view. It is unfortunate that still many Japanese like Imanishi's essays on evolution, probably influenced by his great writing skill and his deep philosophical thinking which I agree to praise.

After finishing the master course, I planned to study in the U.S.A. I loved neutralism so much, and hoped to study with a neutralist. Until that time I read dozens of Dr. Masatoshi Nei's papers, and I was sure that he was a neutralist. In fact, Houston (or Center of Demographic and Population Genetics, Graduate School of Biomedical Sciences, the University of Texas Health Science Center at Houston) was a stronghold of neutralism in 1970's and 1980's. I went to Houston in early September of 1982, and for the first time I met people such as Drs. Wen-Hsiung Li and Ranajit Chakraborty, whose name I knew only by reading their papers.

One of the things I did immediately at Houston was to read Dr. Nei's book (Nei, 1975). That book was in many respects revolutionary, and helped to establish the new field, "molecular population genetics". However, long-term evolution was also within the scope of the book, and he ended that book with the following sentence; Although the mutation we see now is different from that of De Vries and generally minute in effect, it seems to be the primary factor of evolution at

both the molecular and morphological levels. He maintained this view, and explained it in detail in the last chapter of his recent book (Nei, 1987). In fact, he considered to use the term "neo-mutationism" in that book, though somehow dropped that. In any case, I completely agree with this view, because the importance of mutation is a logical consequence of neutralism.

When Dr. Kimura published his book, "The Neutral Theory of Molecular Evolution" (Kimura, 1983), I was anxious to know the content of the book as soon as possible, and borrowed it from Dr. Nei, for Dr. Kimura sent a copy to him. That book is clearly the monument of the neutral theory after 15 years of its birth (Kimura, 1968). Chapter 2 in particular is a detailed yet breathtaking description of the rise and fall of Neo-Darwinism, as the title of the chapter indicates. However, I was not completely satisfied with that book in the following two points. First, I did not agree with his view on the role of the neutral theory on the evolution at the morphological or phenotypic level; he seemed to rely too much on stabilizing selection. There are many phenotypic characters that seem to be impossible to explain solely by stabilizing selection. Here again, mutation should play an important role. Second, there was no mention of philosophical arguments based on the neutral theory. It is possible, however, that Dr. Kimura carefully avoided any philosophical notion based on the neutral theory, as Darwin avoided the discussion on the origin of humans in his book "Origin of Species" (Darwin, 1859).

Dr. Kimura later changed his view on those two aspects (Kimura, 1989, 1990). Now mutations (both gene duplications and point mutations) play important roles in his "four-stage scenario for macroevolution" (Kimura, 1990). Many people will agree that elucidation of evolutionary mechanisms responsible for creating phenotypic or morphological characters is an important subject of evolutionary studies. And I'm sure that the neutral theory will play a central role in phenotypic evolution as it does in "molecular" evolution in the classical sense.

As for the philosophical aspect of the neutral theory, Dr. Kimura himself became Herbert Spencer of neutralism. He coined the term "survival of the luckiest" after "survival of the fittest" (Kimura, 1989). I have a vague feeling that the importance of chance and randomness, as emphasized by Dr. Kimura, may be a key to overcome the mechanistic view of life, which has been the driving force of modern biology in the still continuing battle against vitalism. After vitalism is completely annihilated, as I expect it to be fulfilled soon, we have to face the question, "Then what is life anyway?" In this context, I'm quite convinced that neutralism will become more and more important for understanding the fundamental characteristics of life.

3. A QUESTIONNAIRE SENT TO G. S. J. MEMBERS

While Dr. William Provine of Cornell University was visiting the National

Institute of Genetics in spring of 1991, I came up with the idea of applying a grant to Toyota Foundation with him on the subject of the neutral theory. He kindly accepted my proposal, and I submitted an application with the help of Dr. Yoshio Tateno. We proposed to study reactions and cultural differences toward the neutralism. Although every one but me was skeptical of getting the grant, we were lucky to win it. One year later, Dr. Provine again visited Japan, and three of us (he, I, and Dr. Tateno) fixed the final questionnaire (see the Appendix). During that process, a pre-final version was dispatched via electric mails to about 100 evolutionary biologists, using a network maintained by Dr. Brian Golding. We received scores of responses, and I would like to give one example. Dr. Daniel Dykhuizen sent an E-mail to Dr. William Provine in April 1992. The following is an excerpt of his answer.

> I was still hostile to the neutral theory when I was in Australia in 1974. I remember two things that happened. The first was that I gave a series of talks about the selection-neutral controversy to the Research School of Biological Sciences during which I explained both positions. After I was done, a few people accused me of being a neutralist. By that time I had considerable respect for the theory, but still thought that it was wrong. Later, I tried to explain to Dan Lindsley the neutral theory and why it was wrong. I explained the theory fine but when I explained why it was wrong I ended up blithering-talking on and on without getting anywhere.

[Dr. Dykhuizen did a population genetics experiment using *E. coli* related to the neutral theory (Dykhuizen and Hartl, 1980), and Dr. Kimura coined the term "Dykhuizen-Hartl effect" to call their finding (Kimura, 1983).]

I sent the final questionnaire on the neutral theory to the all members of the Genetics Society of Japan in May 1992, and received responses from 121 persons. Most of them specified their names, and a wide age range is covered; from graduate students to professor emeritus. I appreciate the efforts of those people who responded to our questionnaire. Detailed analysis of those answers as well as interviews will be conducted by Dr. Provine and Ms Tomoko Steen, and I will only briefly introduce some features of the responses.

The followings are a part of answers to question 4—Accepted easily; I felt it reasonable; Eye opening; Touching; Very interesting; Difficult to understand it; My reaction was "neutral"; I do not remember; The neutral theory gives us an insight into the fundamental problems in Biological Science; Inclined to be negative; Very attractive; I was very moved; I was interested in it and impressed profoundly, because I could see another aspect of evolution; Clear and simple; Very convincing; Somewhat difficult, but very fascinating because I doubted the theory of natural selection; Mixed; I was not so surprised by first hearing this theory; Intuitively accepted; Unbelievable. Probably because a majority of persons who responded to our questionnaire reacted positively toward the neutral theory, their answers were often sympathetic with the theory.

Responses to question 6 were more or less as expected. A majority of persons answered that the validity of the neutral theory reduces in the order of a (DNA level), b (protein level), and c (organismal level). There are, however, seven persons who considered the neutral theory's validity at the all three levels. I am in the same opinion with those seven persons.

I insisted on adding question 8 to our questionnaire, but the response was scant. It might have been too early to ask such philosophical questions regarding the neutral theory.

In retrospect, use of questionnaire might not be the best way for our original purpose. Interviews to a restricted number of researchers may be more helpful, as Dr. Provine is continuing to do so. When I gave a short presentation of our result at the Toyota Foundation, Dr. Soichi Iijima, Chief Director, pointed out the same problem. I hope a more thorough research on the historical aspect of neutralism will be conducted in the near future.

4. CONCLUSION

I often envy the contemporary of Charles Darwin, because they could communicate with Darwin himself through real conversation or letters. We, living in the twentieth century, can only read his books and related documents. In the same sense, scientists of the late twenty first century and later will envy us, contemporary of Motoo Kimura.

I thank the Toyota Foundation for supporting us on a historical research of the neutralism.

REFERENCES

Darwin, C. (1859). *The Origin of Species by Means of Natural Selection.* John Murray, London.

Dykhuizen, D. and Hartl, D. L. (1980). Selective neutrality of 6PGD allozymes in *E. coli* and the effects of genetic background. *Genetics* **96**, 801–817.

Imanishi, K. (1976). *What is Evolution*? Kodansha, Tokyo. (In Japanese)

Kimura, M. (1968). Evolutionary rate at the molecular level. *Nature* **217**, 624–626.

Kimura, M. (1976). The stance of the neutral theory in the molecular evolutionary theory and population genetics. *Kagaku* **46**, 528–535. (In Japanese)

Kimura, M. (1983). *The Neutral Theory of Molecular Evolution.* Cambridge University Press, Cambridge.

Kimura, M. (1989). Recent developments of the neutral theory of molecular evolution, and a world view based on the neutral theory. *Jpn. J. Genet.* **64**, 315–334. (In Japanese)

Kimura, M. (1990). The present status of the neutral theory. In: *Population Biology of Genes and Molecules* (eds.: N. Takahata and J. F. Crow), pp. 1–16, Baifukan, Tokyo.

Nei, M. (1975). *Molecular Population Genetics and Evolution.* North-Holland, Amsterdam.

Nei, M. (1987). *Molecular Evolutionary Genetics.* Columbia University Press, New York.

APPENDIX. QUESTIONNAIRE ON THE NEUTRALISM

To: Scientists

We are conducting a study of reactions (positive, negative and indifferent) and cultural differences in reactions to the theory of neutral molecular evolution. Timing is crucial. The neutral theory has been existence for less than a quarter-century, so most people can remember their reactions to the neutralist/selectionist controversies with reasonable clarity.

The study will incorporate answers to the enclosed questionnaire, follow-up questions for some respondents and consequent answers, and selected in-depth interviews. In this initial phase of the study, we have funding sufficient for Japan, USA, and England from Toyota Foundation, but hope to expand the project later.

The questionnaire consists of a necessarily very brief introduction, questions and a literature list. Please feel free to invoke these publications by number in your answers to the questions.

Perhaps you are indifferent to the theory of neutral molecular evolution or know almost nothing about it. We would like to know this fact and want to hear from you even if you do not care to answer the questions.

In addition to regular mail, answers may be returned by Fax or E-mail. The Fax number in Japan is 81–559–75–6040 (from within Japan, 0559–75–6040) and in the USA the Fax number is 1–607–255–8088 (from within USA, 607–255–8088). Our E-mail addresses are:

Provine: wprovine@cornella.bitnet
or wbp@cornella.cit.cornell.edu (for internet)
Saitou: nsaitou@genes.nig.ac.jp
Tateno: ytateno@genes.nig.ac.jp

If you would like to have a copy of our report, please include your return address with your answers. An E-mail return address is ideal for us and saves postage and paper.

We greatly appreciate your help with this brief questionnaire and look forward to your reply. Thank you very much.

Sincerely yours,

William Provine, Naruya Saitou, and Yoshio Tateno

QUESTIONNAIRE

THEORIES OF NEUTRAL MOLECULAR EVOLUTION, 1968–1992

Introduction

February 1993 marks the 25th anniversary of the appearance of Motoo Kimura's 1968 paper in Nature, "Evolutionary rate at the molecular level," presenting his theory of neutral molecular evolution. Suggestions of some degree of neutral

evolution at the protein or DNA level had been frequent since 1962 (including from Sueoka, Freese, Sonneborn, and Jukes), but Kimura's paper was the first detailed presentation of the argument that the vast majority of DNA substitutions was selectively neutral or nearly neutral.

Kimura argued that the calculated rate of evolution over the entire genome extrapolated from available protein data indicated a rate far in excess of the maximum possible rate suggested by J. B. S. Haldane (1957) in his "cost of selection" argument. Kimura deduced the neutral theory from this discrepancy in the rates of evolution utilizing his version of Haldane's argument ("substitutional load"). This paper attracted considerable attention, mostly negative. King (1967), Milkman (1967), and Sved, Reed, and Bodmer (1967) had already proposed truncation selection models countering the argument of Lewontin and Hubby (1966) that heterosis might be insufficient to explain high levels of protein polymorphism. J. Maynard Smith (1968) quickly pointed out that the truncation selection model could also be applied to the rate of evolution showing that substitutional load need not be as great as Haldane and Kimura assumed. Nei (1969) published a paper in early January, 1969, supporting Kimura with the argument that much nonsense DNA existed in vertebrates.

In May, 1969, Jack Lester King and Thomas H. Jukes published in Science their paper on neutral molecular evolution with the provocative title, "Non-Darwinian evolution." Rebuttals to this paper began to appear immediately. King and Jukes (1969) stimulated much opposition not only to itself but also to Kimura (1968), the key critical papers being Richmond (1970) and Clarke (1970), and the chief supporting paper being Arnheim and Taylor (1969).

Kimura followed up his 1968 paper with an extension to "living fossils" but this was refused publication by Nature and appeared in PNAS (Kimura 1969). He and Tomoko Ohta began a series of collaborative papers, perhaps the most visible of which before 1972 was their paper in Nature, "Protein polymorphism as a phase of molecular evolution" (Kimura and Ohta 1971).

Critics of the neutralists focused upon the discrepancies between predictions and measurements. The neutral theory predicted constancy of DNA change by generation, and measured constancy was by year. Measured levels of heterozygosity were constrained within narrow bands and were much less than levels of heterozygosity predicted by the neutral theory. To counter these objections, Ohta invented her "slightly deleterious" or "nearly neutral" model of molecular evolution starting in 1972, bringing it to wide attention with her 1973 Nature paper, "Slightly deleterious mutant substitutions in evolution." Lewontin (1974) published his influential book with detailed evidence for and against the neutral theory, but he argued that to the extent neutral molecular evolution occurred, it was only irrelevant noise and an impediment to the study of adaptive evolution, his primary interest.

All three versions of the neutral theory underwent significant change. Kimur-

a's neutral model in 1979 incorporated his version of Ohta's nearly neutral model. He presented both models, the completely neutral and the nearly neutral, in his 1983 book, The Neutral Theory of Molecular Evolution. The emerging DNA sequence data, however, convinced him about the time of the publication of his book that the completely neutral theory was the only model he wished to develop and defend. By 1985, Kimura no longer incorporated the nearly neutral model, a position in which he has remained consistent to the present.

Jukes has advocated only the completely neutral model. King changed strongly over to Ohta's nearly neutral model by 1975 or 1976. King was much impressed by the criticisms of the completely neutral model but his developing ideas about the neutral theory were cut short by his sudden death in 1983.

Ohta fleshed out her nearly neutral model especially in the years 1972–1976. It gained much strength among evolutionary biologists especially after 1976. From 1976 to 1983, the nearly neutral model was perhaps the most discussed version of the neutral theory. Nei led the opposition to the nearly neutral model during these years, always arguing for the completely neutral version. Ohta's position now is that the nearly neutral model approximates the completely neutral model in organisms with small or intermediate population sizes, but gives qualitatively different results, consistent with observations, in organisms with very large population sizes.

Beginning in the late 1970s, DNA sequence data began to be available, increasing dramatically after about 1983. A sharper distinction between the protein and DNA levels began to appear. The term "molecular evolution" became increasingly vague, requiring a specification of protein or DNA level. Some biologists adopted the neutralist explanation for the majority of evolution at the DNA level, but held a primarily selectionist interpretation at the protein level.

Gillespie (1987, and especially 1991) and Kreitman's laboratory (McDonald and Kreitman 1991) have strongly criticized the neutral interpretation especially at the protein level. Others argue that the great majority of protein evolution is selectively neutral. This is an active area of disagreement at the present time.

A growing number of researchers are focusing upon genomic ecology, finding some parts of the chromosome in which drift dominates, selection in others and investigating experimentally and theoretically the interaction between drift and selection, including hitch-hiking effect (e.g., Begun and Aquadro 1992). This is a very active area of research at the present time.

Optional: Name_____,
Approximate age_____

QUESTIONS

1. What is your current position (e.g. Professor of Genetics) and primary field

of interest (e.g. molecular genetics of yeast)?

2. Where did you do your undergraduate, graduate, and postdoctoral work? What persons during this educational period had the greatest influence upon you with regard to the neutral theory? (e.g, "I worked with Dobzhansky and became a neutralist," or "I worked with Nei and became a selectionist").

3. When and how did you become acquainted with the neutral theory (lecture, seminar, reading, discussion, etc.)? What version of the neutral theory (completely neutral or nearly neutral)? Please refer to the literature list. You may cite publications by number, and certainly feel free to add publications not on the list.

4. What was your initial reaction to the neutral theory? Why?

5. Have your views on the neutral theory changed substantively over time? If so, how and for what reasons (please be as specific as possible, citing publications).

6. What are your views on the neutral theory now, a) at the DNA level, b) at the protein level, c) at the organismal level?

7. We would be keenly interested in any comments you might wish to make on the development of your ideas regarding relationships between neutral theories and a) polymorphism at protein and DNA levels, b) the molecular clock, c) short term vs. long-term evolution, d) silent substitutions in proteins, e) pseudogenes and other non-coding DNA.

8. Do you think the theory of neutral molecular evolution has implications for the controversy between a) "survival of the fittest" vs. "survival of the luckiest" b) determinism vs. indeterminism c) vitalism vs. mechanism d) holism vs. reductionism?

SHORT LITERATURE LIST (in chronological order)

I. Prehistory of the Neutral Theories

1. Sueoka N., 1962. On the genetic basis of variation and heterogeneity of DNA base composition. *Proc. Natl. Acad. Sci. USA* **48**, 582–592.
2. Freese E., 1962. On the evolution of the base composition of DNA. *J. Theor. Biol.* **3**, 82–101.
3. Kimura M. and Crow J. F., 1964. The number of alleles that can be maintained in a finite population. *Genetics* **49**, 725–738.
4. Jukes T., 1966. *Molecules and Evolution.* Columbia University Press, New York.
5. Lewontin R. C. and Hubby J. L., 1966. A molecular approach to the study of genic heterozygosity in natural populations. II. Amount of variation and degree of heterozygosity in natural populations of *Drosophila pseudoobscura*. *Genetics* **54**, 595–609.
6. King J. L., 1967. Continuously distributed factors affecting fitness. *Genetics* **55**, 483–492.
7. Milkman R. D., 1967. Heterosis as a major cause of heterozygosity in nature. *Genetics* **55**, 493–495.
8. Sved, J. A, Reed T. E., and Bodmer W. F., 1967. The number of balanced polymorphisms that can be maintained in a natural population. *Genetics* **55**, 469–481.

II. The Neutral Theories

9. Kimura M., 1968. Evolutionary rate at the molecular level. *Nature* **217**, 224–226.
10. Maynard Smith J., 1968. "Haldane's dilemma" and the rate of evolution. *Nature* **219**, 1114–

1116.
11. Nei M., 1969. Gene duplication and nucleotide substitution in evolution. *Nature* **221**, 40–42.
12. King J. L. and Jukes T. H., 1969. Non-Darwinian evolution. *Science* **164**, 788–798.
13. Kimura M., 1969. The rate of molecular evolution considered from the standpoint of population genetics. *Proc. Natl. Acad. Sci. USA* **63**, 1181–1188.
14. Arnheim N. and Taylor C. E., 1969. Non-Darwinian evolution: Consequences for neutral allelic variation. *Nature* **223**, 900–903.
15. Richmond R. C., 1970. Non-Darwinian evolution: A critique. *Nature* **225**, 1025–1028.
16. Clarke B. C., 1970. Darwinian evolution of proteins. *Science* **168**, 1009–1011.
17. Ohno S., 1970. *Evolution by Gene Duplication.* Springer-Verlag, New York.
18. Kimura M. and Ohta T., 1971. Protein polymorphism as a phase of molecular evolution. *Nature* **229**, 467–469.
19. 1972. *Darwinian, neo-Darwinian and non-Darwinian Evolution.* Proceedings of the Sixth Berkeley Symposium on Mathematical Statistics and Probability, Vol. 5. University of California Press, Berkeley.
20. Wills C., 1973. In defense of naive pan-selectionism. *Am. Nat.* **107**, 23–34.
21. Ohta T., 1973. Slightly deleterious mutant substitutions in evolution. *Nature* **246**, 96–98.
22. Calder N., 1973. *The Life Game.* BBC, London.
23. Lewontin R. C., 1974. *The Genetic Basis of Evolutionary Change.* Columbia University Press, New York.
24. Nei M., 1975. *Molecular Population Genetics and Evolution.* North Holland, Amsterdam.
25. Kimura M. and Milkman R., 1976. Debate on neutral evolution vs. selection. Kimura: "random drift prevails;" Milkman: "selection is the major determinant." *Trends in Biochemical Sciences* **1**, n152–n154.
26. Ohta T., 1976. Role of very slightly deleterious mutations in molecular evolution and polymorphism. *Theor. Pop. Biol.* **10**, 254–275.
27. Kimura M., 1979. Model of effectively neutral mutations in which selective constraint is incorporated. *Proc. Natl. Acad. Sci. USA* **76**, 3440–3444.
28. Kimura M., 1979. The neutral theory of molecular evolution. *Scientific American* **241**, 94–104.
29. Wills C., 1981. *Genetic Variability.* Oxford University Press, Oxford.
30. Kimura M., 1983. *The Neutral Theory of Molecular Evolution.* Cambridge University Press, Cambridge.
31. Ohta T. and Aoki K. eds., 1985. *Population Genetics and Molecular Evolution.* Springer Verlag and Japan Societies Press, Tokyo.
32. Kimura M., 1986. DNA and the Neutral Theory. *Phil. Trans. Royal Soc. Lond. B* **312**, 343–354.
33. Nei M., 1987. *Molecular Evolutionary Genetics.* Columbia University Press, New York.
34. Gillespie J. H., 1987. Molecular evolution and the neutral allele theory. *Oxford Surveys in Evolutionary Biology* **4**, 11–37.
35. Takahata N. and Crow J. F. eds., 1990. *Population Biology of Genes and Molecules.* Baifukan, Tokyo and Princeton University Press, Princeton.
36. Li W.-H. and Graur D., 1991. *Fundamentals of Molecular Evolution.* Sinauer, Sunderland, Mass.
37. McDonald J. H. and Kreitman M., 1991. Adaptive protein evolution at the Adh locus in Drosophila. *Nature* **351**, 652–654.
38. Gillespie J. H., 1991. *Causes of Molecular Evolution.* Oxford University Press, Oxford.
39. Begun D. J. and Aquadro C. F., 1992. Levels of naturally occurring DNA polymorphism correlate with recombination rates in *D. melanogaster*. *Nature* **356**, 519–520.

Note: Fourteen books written in Japanese (list not shown) were added to the above list.