FAT EMBOLISM IN TRAUMA
เอ็มโบลิซึมไขมันในผู้ได้รับอุบัติเหตุ

Visutr Fongsiripaibul
วิสูตร ฟองศิริไพบูลย์

Department of Forensic Medicine, Faculty of Medicine
Siriraj Hospital, Mahidol University
ภาควิชานิติเวชศาสตร์ คณะแพทยศาสตร์ศิริราชพยาบาล
มหาวิทยาลัยมหิดล

ABSTRACT

The study was carried on the cadavers that were autopsied at Siriraj Hospital because of traffic trauma as "Fat Series" and no trauma as "Control Series". Frozen section of lung, kidney, cerebrum, cerebellum and heart were examined for fat emboli by section and staining with oil red O. The results revealed that fat embolism in "Fat Series" and "Control Series" in various organs had statistically significant difference. Lung was the best organ for detecting fat emboli and any lobe of both lungs showed fat emboli in the same pattern. The finding of pulmonary fat embolism in grade 3 or 4 could be a good criteria for confirming antemortem trauma as same as finding only one embolus of kidney, cerebrum and cerebellum or finding grade 2 of cardiac emboli. Femoral and humeral fracture were the most two important causes of fat embolism.

บทคัดย่อ

ได้ศึกษาศพที่ส่งมาชันสูตรที่ภาควิชานิติเวชศาสตร์ คณะแพทยศาสตร์ศิริราชพยาบาล มหาวิทยาลัยมหิดลโดยแบ่งเป็น 2 กลุ่มคือ กลุ่มที่เสียชีวิตเนื่องจากอุบัติเหตุบนท้องถนนและกลุ่มที่เสีย ชีวิตด้วยสาเหตุอื่น โดยการตัดชิ้นเนื้อปอด เนื้อไต เนื้อสมองใหญ่ เนื้อสมองน้อย และกล้ามเนื้อหัวใจ เพื่อตรวจหาเอ็มโบลิซึมไขมัน โดยวิธีแช่แข็ง ตัด และย้อมด้วยน้ำยาออยเรดโอ (oil red O) ผลการศึกษา ปรากฏว่า ทั้ง 2 กลุ่มมีเอ็มโบลิซึมไขมันแตกต่างกันอย่างมีนัยสำคัญ ปอดเป็นอวัยวะที่ใช้ตรวจหา

เอ็มโบลิซึมไขมันได้ดีที่สุดไม่ว่าจะใช้กลีบใดของปอดก็ให้ผลดีเท่าเทียมกัน ข้อบ่งชี้ว่าสาเหตุการตายน่าจะเกิดจากเอ็มโบลิซึมไขมัน คือ การตรวจพบเอ็มโบลิซึมไขมันที่ปอดในระดับ 3 หรือ 4 การตรวจพบเอ็มโบลิซึมไขมันที่ไต เนื้อสมองใหญ่หรือเนื้อสมองน้อย การตรวจพบเอ็มโบลิซึมไขมันที่กล้ามเนื้อหัวใจระดับ 2 และพบว่า กระดูกต้นขาและกระดูกต้นแขนที่หักจะเป็นสาเหตุสำคัญของการเกิดเอ็มโบลิซึมไขมัน

INTRODUCTION

Fat embolism is the blockage of blood vessels by fat globules. The emphasis is on blockage which separates fat embolism from lipaemia in which the plasma fat is a fine emulsion readily passing the smallest capillaries. Embolism by fat has features different from other forms of embolism: it is predominantly a microembolism affecting the minute vessels, especially capillaries and arterioles; the emboli are fluid and deformable. Hence, the vascular occlusion is likely to be temporary.[17]

The present of fat embolism in lung tissue following trauma to bone and soft tissue has been well documented.[1,3,5,6,9,16,18] Fat embolism is thought to originate at the site of trauma[21] from an injured fat depot in the body. Globules of fat released from ruptured fat cell gain access to the circulation through torn venules as a result of a local shift in the differential between extravascular and intravascular pressures. It has been demonstrated that extravascular pressure which is greater than intravascular pressure within the marrow of fractured bones permits entry of fat globules into the circulation.[22] Pulmonary fat embolism happens rapidly,[14,21] usually within a few seconds after injury and a few heart beats are sufficient to bring fat to the lung and even to the systemic circulation.[14] For this reason fat may be found even when death seems to be instantaneous although with sudden death.[14] Pulmonary fat embolism serves as marker for antemortem injury to bone and soft tissue.[12] The amount of fat embolism in the surviving injured cases is proportional to the degree of injury and to the time of survival up to 24 h.[14] Systemic embolism can appear as rapidly as pulmonary embolism and enters to systemic circulation through the lung capillaries. They are most frequently seen in kidney depend on the amount of fat mobilized. The finding of petechial hemorrhages in skin, conjunctiva, mucous membrane and fundus of the eye as well as the finding of fat droplets in sputum or blood can help make a diagnosis of systemic embolism during life.[1,14] Pulmonary fat embolism is rarely found without trauma. Closed chest cardiac massage can result in a small amount of fat embolism depending upon the extend of the injury produced as well as the duration of the survival.[14] Many surgeons believe that it is infrequent or unimportant and a few even doubt about its existence. Others consider it the most frequent cause of death after fracture of long bones. The main objectives in this study are as follows:

1. To compare the degree of fat embolism in lung, kidney, cerebrum, cerebellum and heart between traumatic cases and non-traumatic cases.

2. To find out which organ is the most suitable to indicate fat embolism in traumatic cases.

3. To create criterion for diagnosis of which case fat embolism is possible to be the cause of death.

MATERIALS AND METHODS

Frozen sections of lung, kidney, cerebrum, cerebellum and heart were examined for fat emboli in cadavers that were necropsied at Department of Forensic Medicine, Siriraj Hospital, Mahidol University. The necropsy was divided into two series. The first series named "Fat Series" consisted of 100 cadavers (86 males and 14 females) to be autopsied due to automobile (car, truck, pick up, motor cycle, etc.) accidents. The second series named "Control Series" consisted of 30 persons (24 males and 6 females) dying from drowning, electrocution, and poisoning. All autopsies were personally performed by the same doctor. The history and external wounds of the cadavers were recorded clearly. At each autopsy, specimens from lungs, kidneys, cerebra, cerebella and hearts were collected. Two specimens were collected from each lobe of lung. One specimen was collected from each side of kidney and cerebrum, another was collected from cerebellum and heart. The tissue was fixed in 10% formalin solution and frozen sections were prepared 2 – 3 weeks after the collection. Tissues were cut at 10 – 15 microns intervals. The sections were stained with oil red O for demonstration of fat globules. The examination for fat emboli from the prepared slides under microscope was performed and the grade of each specimen was recorded.

Oil red O in propylene glycol[20]

Fixation	: Formalin solution (10%)
Technic	: Cut frozen sections at 10 – 15 microns
Solutions	: Oil red O in propylene glycol
	Oil red O stain...................0.5g
	Propylene glycol..............100.0 ml

Add a small amount of propylene glycol to the oil red O and mix well. Crush larger pieces. Gradually add the remainder of the propylene glycol, stir regularly. Heat gently until the solution reaches 95°C, do not allow to go over 100°C. Stir while heating. Filter through coarse filter paper while still warm. Allow to stand overnight at room temperature. Filter through Seitz filter under vacuum. If solution becomes turbid, refilter. When using the Seitz filter, put rough surface of filter up.

Staining procedure:
1. Cut frozen sections and store in distilled water.
2. Place sections in absolute propylene glycol for 2 min.
3. Stain in oil red O in propylene glycol for 20 – 30 min.
4. Differentiate in 85% propylene glycol about 1 min.
5. Wash twice in distilled water.

6. Stain in Harris's hematoxylin for a few seconds.

7. Wash twice in distilled water.

8. Differentiate in acid H_2O if overstained.

9. Wash in water.

10. Neutralize in weak ammonia water if differentiate in acid H_2O.

11. Wash twice in distilled water.

12. Mount in glycerin jelly.

Results : Fat   – red

Nuclei   – blue

Grading of fat embolism

The grading of fat embolism was based on the used organs. For pulmonary embolism, about 10 fields of low power field (×10) were counted to assess the mean.

Grade 0 = not found fat embolus

Grade 1 = found 1 – 10 emboli

Grade 2 = found 11 – 30 emboli

Grade 3 = found 31 – 100 emboli

Grade 4 = found more than 100 emboli

Renal embolism was graded according to the proportion of glomeruli containing emboli in 100 glomeruli. Fifty to one hundred glomeruli were examined. Grading was shown below.

Grade 0 = not found fat embolus

Grade 1 = found emboli less than 10% of glomeruli

Grade 2 = found emboli 10 – 30% of glomeruli

Grade 3 = found emboli more than 30% of glomeruli

Grading of cerebrum and cerebellum, the embolism was based on the density of emboli/cm^2 of field in section. Grading was shown below.

Grade 0 = not found fat embolus

Grade 1 = found less than 10 emboli/cm^2

Grade 2 = found equal or more than 10 emboli/cm^2

Grading of cardiac emboli was based on the number of emboli per low power field (×10). About 5 – 10 fields were examined. Grading was shown below.

Grade 0 = not found embolus

Grade 1 = found less than 10 emboli per low power field

Grade 2 = found equal or more than 10 emboli per low power field

RESULTS

There were 130 autopsy cases in this study: 100 cases were organized in "Fat Series" and the rest, 30 cases, were organized in "Control Series." The distribution of age of subjects of both series is shown in Table 1 which reveals mean of both series in nearly the same figure: 30.63 and 31.12. Table 2 shows types of injuries of the subjects in both series, (traffic accidents for "Fat Series" and non-invasive not traffic accidents for "Control Series"). Duration (elapsed time) before death is shown in Table 3.

Statistical analysis of fat embolism in lung, kidney, cerebrum, cerebellum and heart by using Chi square test reveals statistical significant ($p > 0.001$) (Table 4). Table 5 shows the distribution of pulmonary fat embolism in lobes of lung which shows that every lobe can represent total pulmonary fat embolism. The percentage of pulmonary fat embolism in each grade to "Fat Series" is as following: grade 0 = 4%, grade 1 = 5%, grade 2 = 17%, grade 3 = 32%, grade 4 = 35% (Table 5). For "Control Series", the pulmonary fat embolism is found only grade 0, 1 and 2. Total cases of grade 1 and 2 are less than 2%. The number of grade 1 is nearly half of grade 2 (Table 5).

Table 6 shows that the distribution of renal fat embolism from the left side is as the same number as from the right side, either in grade 0, 1, 2, or 3. And only 30% of renal fat embolism is found in traumatic case in "Fat Series" which 22% is grade 1, 15% is grade 2 and 3% is grade 3.

Cerebral fat embolism in "Fat Series" is shown in Table 7 which 32% is grade 1, and 4% of grade 2. Cerebellar fat embolism is occurred only 30% of traumatic cases of "Fat Series" and only 2% is grade 2 (Table 7). In "Control Series", fat embolism in kidney, cerebrum and cerebellum are not found.

For cardiac fat embolism, there is 33% positive for fat; 25% is positive for grade 1 and only 8% is grade 2, as shown in Table 8.

The average grade of fat embolism in lung, kidney cerebrum, cerebellum and heart in "Fat Series" comparing with "Control Series" is shown in Table 9. Statistical analysis, by using t-test, the statistical significance is noted in every organs ($p < 0.05$). Furthermore, this table shows that average grade of pulmonary fat is higher than the others measuring about 2.8.

Many conditions are arranged in "Groups" of the study and analysis. Those are the followings: No Fracture Group, Fracture Group, Long Bone Fracture Group (LBF.), Single Long Bone Fracture Group (Single LBF.), Multiple Long Bone Fracture Group (Multi. LBF.), Not Long Bone Fracture Group (NLBF.), Single Not Long Bone Fracture Group (Single NLBF.), Multiple Not Long Bone Fracture Group (Multi. NLBF.), Only Fracture Skull Group (Only Skull F.), With Vertebral Fracture Group ($\bar{C}$ verteb. F.), With Rib and Clavicle Fracture Group, ($\bar{C}$ Rib & Clav. F.), With Pelvic Fracture Group ($\bar{C}$ Pelvic F.), With Femoral Fracture Group ($\bar{C}$ Femoral F.), With Pelvic Humeral Fracture Group ($\bar{C}$ Humeral F.), With Upper Both Bone

Fracture Group ($\overline{\text{C}}$ upper BB.F.), and With Lower Both Bone Fracture Group ($\overline{\text{C}}$ Lower BB.F.). All are present in Table 10. Fracture femur is the most important cause of fat embolism in every studied organ. Fracture vertebra is the less important cause of fat embolism in fracture group of traumatic cases.

DISCUSSION

Pulmonary fat embolism is a very common necropsy finding in injured subjects, especially in those with fractures. The fat globules in the pulmonary vessels were frequently causes of unexplained death. Pulmonary fat emboli are found in branches of the pulmonary artery, in the arterioles, and in the alveolar capillary bed but free in the alveolar space. They vary in size most are 20 – 40 microns in diameter, but they range up to 100 microns or more presumably because multiple smaller droplets have fused together. Within capillaries the emboli are often found as elongated cylindical forms or compressed sausage-like bodies of branching at T and Y capillary or arteriolar junctions.[17] Pulmonary fat embolism happens rapidly[21] usually within a few seconds after injury and a few heart beats are sufficient to bring fat to the lung and even to the systemic circulation.[14] In animal, Busch[4] showed that lung emboli appeared within minutes of a fracture or marrow trauma; Olbrycht[13] found that lung emboli appeared with seconds of crushing the extremities. The speed of lung embolism is quick provided that there is more than a feeble circulation. So, it can gave medicolegal significance.

Pulmonary fat embolism in "Fat Series" is different from "Control Series" with statistical significance ($P > 0.001$ by using Chi square test). The mean grade of pulmonary embolism in this study is 2.84, comparing with 0.8 of "Control Series", which has statistical significance ($p < 0.05$ by using t-test) (Table 9). This means that pulmonary fat embolism is a criteria for indicating antemortem change in traumatic cases (Table 4). The study which included lobe of lung (right upper lobe (RUL), right middle lobe (RML), right lower lobe (RLL), left upper lobe (LUL) and left lower lobe (LLL)) is suitable for collection as the sample to examine fat emboli; and reveals that any lobe can be effectively used as specimen for detecting fat emboli (Table 5). The important remark is that "Control Series" is not found pulmonary fat embolism in grade 3 and 4. So, the conclusion is that pulmonary fat emboli in grade 3 and 4 can use for the criteria indicating antemortem blunt force injury.

Fracture femur which is the most important cause of pulmonary embolism, demonstrating by means of the grade of "With Femoral Fracture" had the highest score (3.4316) and vertebral fracture is the less important cause of pulmonary embolism with mean grade of 2.2714 (Table 10).

Kidneys are very vascular. All the renal blood supply passes through the glomerular capillaries. For this reason, examination of a frozen section of the kidney at necropsy is the most reliable single test of systemic embolism.[17] Renal fat embolism can produce the condition of uremia and anuria.[8] The microscopic feature is the presence of fat emboli in

glomerular capillaries, parts of which are distended by sausage-shaped, oval or round emboli.

The study of renal fat embolism, comparing between "Fat Series" and "Control Series", reveals statistical significance ($\chi^2 = 14.9087$, p > 0.001) (Table 4). Right and left kidneys can be found fat embolism in the same pattern. Because of no case in "Control Series" which had positive fat emboli, so renal fat embolism can be used for a criteria for diagnosis of traumatic cause of death. The average grade of renal fat embolism is as follows: grade 0 = 70%, grade 1 = 22%, grade 2 = 5%, grade 3 = 3%. Mean of grade of "Fat Series" is 0.395 which is statistically significant after comparing to "zero" of "Control Series" (Table 9). Mean of grade of renal fat embolism in almost all of the conditions is less than 1, except the condition of "With Humeral Fracture Group". This shows that fracture humerus is the most important cause of systemic fat embolism (Table 10).

Cerebral fat embolism has been found at autopsy in patients who had clinical evidence of fat embolism and who died after massive trauma.[10] It is seldom appearing as a pure isolated cerebral form. It can precede, parallel or follow up the appearance of pulmonary involvement.[2,11] The brain often appears edematous. Microscopic finding reveals a profusion of fat emboli, as well as some areas of necrosis and hemorrhage, in the grey mater and deep nuclei.[19]

The study of cerebral fat embolism comparing between "Fat Series" and "Control Series" reveals statistical significance ($\chi^2 = 15.5064$, p > 0.001) (Table 4). Positive finding for cerebral fat embolism is only 36% of cases and only 4 are in grade 2 (Table 7). No case in "Control Series" has positive for cerebral fat emboli, so cerebral fat embolism can be used to a criteria for diagnosis of traumatic cause before death. Average grade of cerebral fat embolism comparing with "Control Series" has significant difference (p < 0.05, by using t-test). Cerebellar fat embolism nearly behaves the same pattern as cerebral fat embolism.

Myocardial embolism receives emboli via the coronary arteries. The symptom of cardiac embolism may be confused with fulminanting type of pulmonary fat embolism.[7,15] In frozen section fat emboli are found within the network of capillaries between the muscles fibers usually as sausage-shaped or narrow, elongated, deformed tubular structure. There are free hemorrhage around and between the muscle fibers, or focal areas of fatty change in fibers adjacent to the emboli or both hemorrhage and fatty change.[17] The cases with many myocardial emboli are also those with numerous emboli in the brain and elsewhere. The significance of cardiac emboli is difficult to assess.

The study of cardiac fat embolism comparing between "Fat Series" and "Control Series", reveals statistical significance ($\chi^2 = 9.0361$, p > 0.005) (Table 4). Positive cardiac fat embolism in "Fat Series" is 33% (25% of grade 1 and 8% of grade 2). There is only one case (3.3%) of "Control Series" with positive grade 1 (Table 8). So, finding of cardiac fat embolism indicates antemortem traumatic change. Mean of the grade of "Fat Series" and "Control Series" are 0.41 and 0.0333, respectively, which had statistical difference (p < 0.05, by using t-test) (Table 9). Mean of the grade of cardiac fat embolism in every conditions of "Fat Series" are less than 1 (Table 10).

CONCLUSION

Frozen section of lung, kidney, cerebrum, cerebellum and heart were examined for fat embolism in cadavers to be necropsied due to traffic trauma (Fat Series) and no trauma (Control Series). The two series have statistically significant difference in various organs. Lung is the best organ for detecting fat emboli and any lobe of lung gives the same pattern of emboli. Fracture femur is the most important cause of fat embolism. The criterion for diagnosis of antemortem fat embolism are; (1) the finding of grade 3 or 4 of pulmonary fat embolism, (2) the finding of grade 2 of cardiac fat embolism or (3) the finding of one of renal, cerebral or cerebellar fat embolism. The three above criteria can be used as the cause of death if there are some clinical signs and symptoms of fat embolism syndrome and no other causes of death.

ACKNOWLEDGEMENTS

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Table 1.   Age distribution of subjects

Age	Fat Series	Control Series
< 10	1	3
11 – 20	28	6
21 – 30	38	9
31 – 40	8	3
41 – 50	13	5
51 – 60	3	3
> 60	8	1
Mean	31.12	30.63

Table 2.   Types of injury of "Fat Series" and "Control Series"

Types of injury	Fat Series	Control Series
Motor cycle accident	54	—
Car accident	22	—
Pick up accident	4	—
Train accident	3	—
Bus accident	4	—
Truck accident	10	—
Bicycle accident	3	—
Electrocution	—	9
Drowning	—	18
Poisoning	—	3
Total	100	30

Table 3.   Duration before death

Times (h)	Fat Series	Control Series
At once (0)	38	25
$>$ 0 – 1	20	1
$>$ 1 – 24	14	3
$>$ 24 – 48	7	0
$>$ 48 – 72	10	0
$>$ 72	11	1
Total	100	30

Table 4.   Comparison of fat embolism between two series in various organs

Organs	Fat Series	Control Series	χ^2	P
Lung	99	3	102.4621	$>$ 0.001
Kidney	39	0	14.9087	$>$ 0.001
Cerebrum	40	0	15.5064	$>$ 0.001
Cerebellum	30	0	8.3925	$>$ 0.005
Heart	33	1	9.0361	$>$ 0.005

Table 5. Pulmonary fat embolism in "Fat Series" and "Control Series"

Organ	Grade of pulmonary fat embolism					Total
	0	1	2	3	4	
Fat Series						
RUL	4	4	21	35	36	100
RML	3	3	14	41	37	100
RLL	6	3	12	41	38	100
LUL	2	10	22	36	30	100
LLL	3	7	17	40	33	100
Average	4	5	17	39	35	100
Control Series						
RUL	27	2	1	0	0	30
RML	28	1	1	0	0	30
RLL	27	2	1	0	0	30
LUL	29	1	0	0	0	30
LLL	29	1	0	0	0	30
Average	28	1.5	0.5	0	0	30

Table 6. Renal fat embolism in "Fat Series"

Kidney	Grade				Total
	0	1	2	3	
Right	67	25	6	2	100
Left	74	19	4	3	100
Average	70.5	22	5	2.5	100

Table 7. Cerebral and cerebellar fat embolism in "Fat Series"

Organs		Grade			Total
		0	1	2	
Cerebrum	Right	61	35	4	100
	Left	67	28	5	100
	Average	64	31.5	4.5	100
Cerebellum		70	28	2	100

Table 8. Cardiac fat embolism

Organ	Grade			Total
	0	1	2	
Heart (Fat Series)	67	25	8	100
Heart (Control Series)	29	1	0	30

Table 9. Mean of the grade of fat embolism in various organs

Organs	Fat Series (100 cases)	Control Series (30 cases)	t	P
Lung	2.84	0.0867	24.2820	< 0.05
Kidney	0.395	0	6.0523	< 0.05
Cerebrum	0.415	0	7.4729	< 0.05
Cerebellum	0.32	0	6.2410	< 0.05
Heart	0.41	0.0333	5.2395	< 0.05

Table 10. Mean of the grade of fat embolism of various organs in different conditions

Condition	No. of cases	Mean of the grade of				
		Lung	Kidney	Cerebrum	Cere-bellum	Heart
Fat Series	100	2.84	0.395	0.4150	0.32	0.41
Control Series	30	0.0867	0	0	0	0.0333
No Fracture	2	2.90	0.25	0.50	0	0.50
Fracture	98	2.8429	0.398	0.4031	0.3265	0.4082
LBF.	45	3.1711	0.6333	0.5333	0.4667	0.6222
Single LBF.	20	3.125	0.6750	0.5250	0.5500	0.550
Multi. LBF.	25	3.2080	0.6000	0.5400	0.4000	0.680
NLBF.	53	2.5642	0.1981	0.2925	0.2075	0.2264
Single NLBF.	30	2.5167	0.1833	0.3833	0.20	0.2333
Multi. NLBF.	23	2.6261	0.2174	0.1739	0.2174	0.2174
Only Skull F.	24	2.6375	0.2083	0.3750	0.2500	0.2083
C̄ verteb. F.	7	2.2714	0.0714	0	0	0
C̄ Rib & Clav.F.	37	2.8784	0.4459	0.3919	0.3514	0.3514
C̄ Pelvic F.	13	3.0767	0.4231	0.3846	0.3846	0.6154
C̄ Femoral F.	19	3.4316	0.8421	0.6842	0.5789	0.7368
C̄ Humeral F.	14	3.3143	1.0357	0.8571	0.7143	0.9286
C̄ Upper BB.F.	6	2.8500	0.8333	0.8333	0.5000	0.6667
C̄ Lower BB.F.	21	3.1952	0.5952	0.5238	0.3810	0.7619